A STUDY OF PHYSIOLOGICAL AND PHARMACOLOGICAL INTERVENTIONS ON MYOCARDIAL CONTRACTILITY

BY

KATHLEEN ANN KANE.

THESIS SUBMITTED FOR THE DEGREE OF DOCTOR OF PHILOSOPHY, UNIVERSITY OF EDINBURGH, 1974.

ABSTRACT OF THESIS

Name of Candidate Kathleen A. Kane

Address .....

Degree Ph.D. *Date* 11th September, 1974.

Title of Thesis A study of physiological and pharmacological interventions on myocardial contractility.

In view of the recent work on isolated cardiac muscle which has shown that situations can occur in which the force and velocity of contraction are changed in different directions, we have questioned the usefulness of the search for a single index of contractility related to the rate of rise of tension, a trend which dominates studies of the heart in vivo. We have attempted to show, in the heart in vivo, that situations do occur in which the duration of contraction can determine the force of contraction or in which force and velocity of contraction are changed in different directions such that purely a change in the velocity dependant aspects of contraction would not describe the inotropic intervention.

Dog hearts have been prepared in situ so that heart rate (HR), left ventricular end diastolic pressure (LVEDP) and mean aortic pressure (MAP) can be controlled separately during computation of left ventricular $\frac{dP}{dt}$ max and external stroke work (SW). Progressive increases in HR consistently raised $\frac{dP}{dt}$ max over a wide range, and consistently lowered SW except at low rates. Progressive increases in LVEDP or MAP consistently raised both $\frac{dP}{dt}$ max and SW. Infusion of noradrenaline consistently raised both $\frac{dP}{dt}$ max and SW, except at very high HR when only $\frac{dP}{dt}$ max was consistently raised.

We have investigated the haemodynamic effects of sotalol and bretylium which have been reported to increase the force of contraction by prolonging the duration of the contraction in isolated cardiac muscle. We were unable to show that these drugs produced divergent effects on $\frac{dP}{dt}$ max and SW, although sotalol but not bretylium did prolong the duration of contraction.

We also investigated the cardiovascular effects of prostaglandins C and E and found that both prostaglandins have a positive inotropic effect in the dog, but not in the cat.

Our results lead us to question the validity of equating changes in $\frac{dP}{dt}$ max with changes in indices of performance of the heart as a pump under abnormal conditions and in the assessment of inotropic agents.

SUMMARY

In view of the recent work on isolated cardiac muscle which has shown that situations can occur in which the force and velocity of contraction are changed in different directions, we have questioned the usefulness of the search for a single index of contractility related to the rate of rise of tension, a trend which dominates studies of the heart in vivo. We have attempted to show, in the heart in vivo, that situations do occur in which the duration of contraction can determine the force of contraction or in which force and velocity of contraction are changed in different directions such that purely a change in the velocity dependant aspects of contraction would not describe the inotropic intervention.

Dog hearts have been prepared in situ so that heart rate (HR), left ventricular end diastolic pressure (LVEDP) and mean aortic pressure (MAP) can be controlled separately during computation of left ventricular $\frac{dP}{dt}$ max and external stroke work (SW). Progressive increases in HR consistently raised $\frac{dP}{dt}$ max over a wide range, and consistently lowered SW except at low rates. Progressive increases in LVEDP or MAP consistently raised both $\frac{dP}{dt}$ max and SW. Infusion of noradrenaline consistently raised both $\frac{dP}{dt}$ max and SW, except at very high HR when only $\frac{dP}{dt}$ max was consistently raised.

We have investigated the haemodynamic effects of sotalol and bretylium which have been reported to increase the force of contraction

Summary Continued:

by prolonging the duration of the contraction in isolated cardiac muscle. We were unable to show that these drugs produced divergent effects on $\frac{dP}{dt}$ max and SW, although sotalol but not bretylium did prolong the duration of contraction.

We also investigated the cardiovascular effects of prostaglandins C and E and found that both prostaglandins have a positive inotropic effect in the dog, but not in the cat.

Our results lead us to question the validity of equating changes in $\frac{dP}{dt}$ max with changes in indices of performance of the heart as a pump under abnormal conditions and in the assessment of inotropic agents.

CONTENTS.

INTRODUCTION

The effect of initial muscle length on the contraction of the muscle is part of the fundamental background of muscle physiology, and was first observed in skeletal muscle (Blix 1895). The effect was confirmed in cardiac muscle by Frank (1895). Using an isolated frog heart preparation which was contracting isometrically, he showed that ''the peaks of the isometric curves rise with increasing initial tension (filling)''. Although Frank implies that end diastolic filling is the important factor, his experiments were designed to measure only end diastolic tension, and so it remained for Starling and his co-workers to specify initial muscle length as the prime determinant of work done during the following contraction. Starling (1918) showed in a dog heart-lung preparation that with a moderate increase in inflow or arterial resistance there was an increase in end diastolic muscle length, but little or no rise in end diastolic pressure. Yet stroke work (when aortic pressure was raised) and stroke volume (when inflow was increased) were invariably increased. These findings led Starling to state that 'the Law of the heart is thus the same as the Law of muscular tissue, generally that the energy of contraction however measured, is a function of the length of the muscle fibre. This length-energy relationship dominated the field of cardiac function for the next thirty years, in which time the inability of Starling's Law to explain the cardiovascular phenomena associated with exercise led Hamilton (1955) and Rushmer

(1955) and other workers to question its importance in the intact animal. Starling himself, however, had considered that the length-energy relationship was not the sole mechanism responsible for the increased output in exercise. ''But the effect of throwing this organ (heart) into the circle of control by the central nervous system is that it is kept in rest or activity in an equable condition and the dilatation, which was so marked a condition of its reaction when isolated, is reduced to such small dimensions in the heart reined in and controlled by the cardiac centres, and helped by the correlated changes in other organs, that it becomes imperceptible in the intact animal, and is not revealed for instance, by any radiographic study of the heart during exercise.'' (Starling 1920).

The paradox of a decrease in end diastolic volume (EDV) and an increase in stroke work (SW) observed in exercise was explained by Sarnoff & Berglund (1954). They introduced the concept of a family of ventricular function curves but attributed the idea to J.W. Dow. In their experiments, mean atrial pressure was used as an indication of the EDV of the ventricle and external SW (measured by multiplying stroke volume by mean arterial pressure) as an index of the force of contraction of the left ventricle. They established the ventricular function curve which expresses the increases in external SW associated with increments in initial muscle length according to Starlings Law, and showed that when the contractile state of the heart is altered, by injection of adrenaline, the ventricular function curve is shifted to the left,

i.e. an increase in the contractile state of the heart is associated with an increase in external SW at any given muscle length. This concept explained why in exercise Starling's Law still applied although SW was increased while filling pressure remained the same or decreased. In later experiments Sarnoff et al (1960a) used the concept of a family of ventricular function curves to quantitate the inotropic response of the heart to cardiac sympathetic nerve stimulation. In these experiments an increase in ventricular contractility was defined as an increase in external SW at any given left ventricular end diastolic pressure (LVEDP). These experiments have been criticised, because it has been shown that changes in heart rate and in mean arterial pressure (MAP) can also influence the external SW.

An increase in the amplitude of contraction resulting from an increase in the frequency of contraction was first noted by Bowditch (1871) in the frog heart. This effect was confirmed in the intact animal by Rosenblueth et al (1959a) and by Sarnoff et al (1960b). Rosenblueth et al showed, that in the perfused dog heart contracting isometrically, when the frequency of stimulation was increased, the ventricular contractions gradually increased to become stronger than the control contractions. The return to the control rate produced a gradual decline of the contraction amplitude to the original level. The existance of this staircase effect, as it has become known, necessitates the control of heart rate in studies on the quantification of

myocardial contractility.

Two distinct mechanisms by which a change in MAP can alter the amount of work done in a contraction have been reported. From the mechanical properties of muscle, one would expect that the work performed in a contraction is governed partly by the resistance to shortening (afterload) that is offered by the system in which the muscle is working. External work must be zero both when external resistance is zero and when it is too great for shortening, with a maximum in between. Changes in afterload can, therefore, produce either a decrease, if the afterload is increased above the optimal one, or an increase in the work performed in a contraction from a given muscle length. In the intact animal, MAP approximates to the afterload in an isolated muscle; changes in MAP may therefore produce changes in the SW perfomed from a given EDV even although myocardial contractility is constant. This effect was confirmed in the cat heart in situ by Sonnenblick and Downing (1963). In their preparation, the heart was paced and reflex activity was abolished by ligation of the brachiocephalic artery. They showed that at a given LVEDP, stroke volume was not affected appreciably by changes in MAP over a wide physiological range. It follows that at any given LVEDP, SW is a function of MAP.

The other effect which has been reported when MAP is changed is that described as the Anrep effect. Anrep (1912) and Knowleton and Starling (1912) first observed that if the aortic pressure of the heart-lung preparation was increased suddenly, the diastolic

volume first rose, resulting in an increased output. After a number of beats the same output was maintained although the diastolic volume had returned to its control level. A gradual increase in myocardial contractility must have occurred. The increased contractility persists as long as the aortic pressure remains raised. The mechanism responsible for the Anrep effect is not known. The Anrep effect is to be distinguished from the other mechanism described by which an increase in MAP may produce an increase in SW at constant LVEDP, but which need not imply a change in the contractile state of the heart.

As variables, such as heart rate and MAP, have been shown to affect the energy of contraction, the ventricular function curve cannot be regarded as a unique representation of the contractile state of the heart. Although Sarnoff (1960b), himself investigated the effects of MAP on SW, he used external SW, as an index of myocardial contractility in experiments in which MAP was subject to change. Under these conditions, one cannot assess to what extent a change in SW was due to a change in MAP, or to a direct effect of adrenaline on the contractile elements of the heart. These limitations in the use of SW to describe changes in the contractile state of the heart led to a search for an index of contractility which was independant of changes in heart rate, LVEDP and MAP.

Around the same time, it was realised that a more precise definition of the nature of cardiac contractility, in terms of the concepts developed by A.V. Hill for skeletal muscle, was

necessary as a basis for the appreciation of cardiac dynamics: particularly in order to formulate in fundamental terms changes in myocardial contractility and the mechanisms by which these changes were accomplished. The essence of the concepts developed by A. V. Hill (1938) for skeletal muscle is that a muscle behaves as if it were composed of three components.

1. An active contractile element (CE) which can develop tension or shorten according to a particular force-velocity relationship.
2. A series elastic element mechanically in series with the contractile element.
3. A parallel elastic element mechanically in parallel with the CE.

When the CE is actively shortening or developing force, the muscle is said to be in the active state. Using this model, it is clear that two fundamental parameters determine the mechanical response of the muscle. One relating directly to force development and the other relating to the velocity of shortening. Under any given circumstance, the velocity of shortening of the CE is uniquely determined by the tension in the muscle. As the tension is increased, the velocity of shortening is decreased, according to the equation formulated by Hill.

$$(P + a)(v + b) = (Po + a)b = \text{Constant}$$

where P is the load lifted: v is the velocity of shortening: Po is the maximum isometric tension: a & b are constants.

The graph of velocity of shortening as a function of tension is generally described as being hyperbolic for skeletal muscle, and it intersects the axes at finite values of force and veloctiy. The unique properties of the force-velocity relation hold only under conditions where P and V are measured at a constant Po i.e. at a muscle length where Po is not length dependant and the active state intensity is not changing with time. For this reason the force veloctiy relation is most easily studied and is most useful for quantification of the CE in skeletal muscle in which Po is constant when the muscle is maintained in a steady state of full activity by tetanic stimulation. In a tetanic isometric contraction, the tension developed, Po, depends only on the maximum intensity of the active state and not on the duration of the active state or the intrinsic speed of the muscle. The force intercept of the instantaneous force-velocity curve, therefore, corresponds to the intensity of the active state, which provides a useful index of the degree of activation of the CE at that instant. Similarly the intercept on the velocity axis will provide a measure of the maximum speed per unit muscle length at which the muscle shortens under zero load. An inotropic change can therefore be defined as a change in the force-velocity relationship of the muscle, when the measurements of force and velocity are made at constant CE length and time after excitation. It has been shown, however, that in cardiac muscle, it is not possible in an afterloaded isotonic contraction to measure the force-velocity relationship of the CE

within the necessary conditions of constant CE length and time after excitation (Brady 1968). This is because skeletal muscle and cardiac muscle differ in four important respects..

1. Cardiac muscle cannot be tetanized. Therefore, Po, the maximum force generating capacity of the muscle cannot be found directly, but only by extrapolation of the force-velocity curve. This is not accurate, particularly since peak isometric twitch tension in cardiac muscle can be modified greatly by factors such as frequency of contraction and humoral agents.
2. In cardiac muscle the development of contractile activity is relatively slow compared with skeletal muscle. In afterloaded contractions, therefore, the instantaneous force-velocity relation measured at the time each load is lifted is different from that at any other load, and the velocity at which high loads are lifted may not be maximal intially.
3. In skeletal muscle a range of muscle lengths near body length can be selected for study in which the tetanic isometric force developed is relatively constant. Thus varying degrees of internal shortening occurring with different afterloads do not appreciably alter the driving force of the contraction. In cardiac muscle, changes in muscle lengths close to body length cause changes in the peak isometric twitch tension.
4. At this range of muscle lengths little resting tension exists in skeletal muscle. The load lifted by the CE is constant and is thus not affected by the transfer of resting tension to

the CE as muscle shortening occurs. In cardiac muscle there is a considerable resting tension at muscle lengths where maximum tension is developed, and therefore, the load on the CE, as the muscle shortens is not constant.

The force-velocity relations in cardiac muscle abound with uncertainties. Since Po becomes a time dependant function in cardiac muscle, the available energy in the system at any moment is time dependent. Such a system is no longer a unique function of force and velocity. Although the basic processes of actomyosin association in cardiac and skeletal muscles are similar: the dependance on time and length, and the influence of resting tension on cardiac CE force-velocity curves makes the formulation of the basic principles governing cardiac mechanics very difficult using classical techniques developed for skeletal muscle. Furthermore the concept of active state which was developed for skeletal muscle loses much of its advantage when applied to cardiac muscle. In skeletal muscle, the intensity of the active state reaches a plateau which can be maintained by repetitive stimulation. Various properties of the muscle are studied in the state of full activity and it is possible to ascribe changes in twitch tension either to a change in the duration or the intensity of the fully active state. In the case of cardiac muscle there is not a plateau of activity and so it is less meaningful to consider any property of heart muscle in terms of fully active state.

Such analysis, however, has been applied to cardiac muscle

in an attempt to define inotropic changes in terms of the fundamental properties of muscle. Abott and Mommaerts (1959) in the first attempt to analyse some of the fundamental mechanical characteristics of cardiac contractility, used the force-velocity relationship in cat papillary muscle to show that the positive inotropic effect, produced by an increase in frequency of contraction, was associated with an increase in the velocity of contraction as well as an increase in the force of contraction. These studies were extended by Sonnenblick (1962) who extrapolated the force-velocity curves and related inotropic changes to changes in the maximum velocity of shortening, Vmax, the intercept on the velocity axis. He reported that an increase in initial muscle length caused an increase in rate of development of force, an increased Po, but no change in Vmax. He concluded that, since Vmax was independent of changes in initial muscle length, Vmax could be used to quantitate the contractile state of the heart. This idea was further supported by his results that inotropic interventions, such as increased frequency of contraction or the addition of noradrenaline was characterised by an increase in Vmax with a variable effect on Po. The force-velocity curves in both these studies were plotted from the initial velocity of shortening in contractions lifting various afterloads. For the reasons stated above, ideally the relation of force to velocity should be determined at a given time after excitation. It is possible, therefore, that changes in the shape of Sonnenblick's curves could be consistant with changes in the time course

of the contraction.

The results of these first experiments in cardiac muscle led to the realisation that the rate of development of force as well as the force of contraction required consideration in connection with concepts of contractility. Although the experimental difficulties involved in measuring Vmax negate its value as a measurement of contractile element velocity, it does reflect changes in the rate of development of tension and as such is useful. In the intact heart, force in the left ventricular wall is related to left ventricular pressure, P, though ventricular dimensions. Hefner et al (1962) showed experimentally that:-

$$F = Pa. \qquad \text{equation 1.}$$

where F = Force tending to pull the ventricle into two halves

P = left ventricular pressure.

a = cross sectional area of the cavity in the plane of division of the ventricle into two halves.

The rate of development of force can, therefore, be related to the rate of development of pressure in the ventricle, as follows, differentiating equation 1 with respect to time.

$$\frac{dF}{dt} = \frac{dP}{dt}\, a + \frac{da}{dt}\, P$$

If the analysis is confined to the isovolumic contraction period, and the assumption is made that the contraction is isometric,

i.e. $\frac{da}{dt} = 0$ then,

$$\frac{dP}{dt} = \frac{dF/dt}{a} \qquad \text{equation 2.}$$

The maximum rate of rise of ventricular pressure ($\frac{dP}{dt}$ max) during the isovolumic phase can be used as an indirect measure of the maximum rate of ventricular work. During an isovolumic contraction, as the CE shortens at a particular velocity, it stretches the series elastic element. The rate of development of pressure in the ventricle is, therefore determined by the velocity of contraction of the CE, the elasticity of the series elastic element and the size of the ventricle. Since no inotropic interventions have been shown to alter the properties of the series elastic element, it can usually be assumed that $\frac{dP}{dt}$ max can be used as an index of a change in the velocity of the CE. An increase in initial muscle length increases $\frac{dF}{dt}$ (Sonnenblick 1962). An increase in EDV induces an increase in initial muscle length and an increase in a, in equation 2. The effect of an increase in heart size on $\frac{dP}{dt}$ max will, therefore, depend on the balance between the increase in $\frac{dF}{dt}$ and the increase in a. If the changes do not balance, $\frac{dP}{dt}$ max will be sensitive to changes in heart size. $\frac{dP}{dt}$ max ought to be insensitive to changes in MAP, if it is measured within the isovolumic phase.

As early as 1914, Wiggers showed that adrenaline increased the mean rate of rise of pressure in the ventricle. He also reports that when end diastolic filling pressure was raised by increasing venous return to the heart, the rate of rise of pressure was also increased. Since this report, there has been much disagreement in the literature on the question of

the independence of $\frac{dP}{dt}$ max on LVEDP and MAP. Reeves et al (1960) showed that in the anaesthetized dog, changes in $\frac{dP}{dt}$ max, induced by noradrenaline had a significant correlation with changes in peak ventricular pressure and changes in contractile force measured with a strain gauge arch. $\frac{dP}{dt}$ max also varied with changes in ventricular end diastolic pressure. In these experiments, MAP was allowed to change freely. It has since been shown that $\frac{dP}{dt}$ max does vary with changes in MAP (Clancy et al 1968). For this reason, one cannot conclude from these experiments that $\frac{dP}{dt}$ max is independent of LVEDP. Wallace(1963), however, observed the effect of changing either HR, MAP or LVEDP on $\frac{dP}{dt}$ max while the other two variables were held constant. He concluded that an increase in LVEDP did produce an increase in $\frac{dP}{dt}$ max.

These results impose the same limitations on $\frac{dP}{dt}$ max when it is used as an index of myocardial contractility as are imposed on SW. In spite of these limitations, $\frac{dP}{dt}$ max has become a popular index of contractility, particularly as it is very sensitive to the inotropic effect of catecholamines. The effect of catecholamines on $\frac{dP}{dt}$ max was first established by Gleason and Braunwald (1962) in human subjects. Large increases in $\frac{dP}{dt}$ max were recorded in response to intravenous injections of isoprenaline and noradrenaline. These results were confirmed by Franklin et al (1962) who showed that in anaesthetized dogs, marked increases in $\frac{dP}{dt}$ max were obtained during changes in HR, exercise and upon infusion of isoprenaline.

In a sense, it was unfortunate that the inotropic effect of catecholamines is characterized primarily by such a marked effect on the rate of contraction, because this led to the idea that inotropic changes were determined by a single factor, the change in Vmax. This in turn led to a search for a single index of contractility occurring in the isometric phase of contraction which reflected a change in Vmax. Siegel and Sonnenblick (1963) introduced the index $\frac{dP}{dt}\Big|IIT$: where IIT is a constant fraction of the integrated systolic isometric tension based on the time to $\frac{dP}{dt}$ max. Although $\frac{dP}{dt}$ max and IIT both increase as EDV is increased, the ratio $\frac{dP}{dt}\Big|IIT$ was claimed to be independent of changes in muscle length. In a later study changes in this index were compared with changes in the ventricular function curve (Siegel et al 1964). Sympathetic nerve stimulation via the stellate ganglion at constant HR, produced a marked shift to the left in the ventricular function curve. This was associated with an increase in $\frac{dP}{dt}$ max and an increase in IIT, but because the increase in $\frac{dP}{dt}$ was proportionally greater than the increase in IIT, an increase in the ratio $\frac{dP}{dt}\Big|IIT$ also occurred. This increase in $\frac{dP}{dt}\Big|IIT$ was also independent of changes in LVEDP. The effect of a small increase in HR was also observed. No detectable change in the ventricular function curve was obtained but because there was an increase in $\frac{dP}{dt}$ max with virtually no change in IIT, the index $\frac{dP}{dt}\Big|IIT$ increased. From these results, it was concluded that changes in contractility were characterized by a change in

the velocity dependant aspects of contraction which may or may not be associated with changes in the force of contraction. The ratio $\frac{dP}{dt}\Big|_{IIT}$ was claimed to be a more reliable index of myocardial contractility than the ventricular function curve, because, a) $\frac{dP}{dt}\Big|_{IIT}$ is independent of changes in LVEDP and thus enables distinction between the Starling mechanism and true inotropic changes.

b) $\frac{dP}{dt}\Big|_{IIT}$ reflects changes in the velocity of contraction better than a ventricular function curve.

Attempts have also been made to derive force-velocity relations from the isovolumic phase of contraction in the intact heart. In addition to the theoretical objections already described, the measurement of both the velocity and the load in the intact heart requires the consideration of several additional factors that are difficult to quantitate precisely. For example; the assumption of uniform wall thickness and that all fibres run circumferentially and contract synchronously, the use of a spherical model for the left ventricle, and the assumption that absolute initial volume of the ventricle may be derived from the end diastolic pressure in the ventricle by using a pressure volume curve. The velocity of the CE is calculated by assuming a two compartment model of heart muscle, in which the CE, as it shortens during the isovolumic contraction of the muscle, stretches a series elastic component. The velocity of the series elastic element extension $\left(\frac{dl}{dt}\right)$ and therefore the velocity of CE shortening is considered to be directly related to the rate of force

development $\left(\frac{dF}{dt}\right)$ and inversely related to the stiffness of the series elastic element. $\left(\frac{dF}{dl}\right)$

$$\text{i.e. } \frac{dl}{dt} = \frac{dF/dt}{dF/dl} \qquad \text{equation 3.}$$

The stress-strain relation of the series elastic element, measured experimentally in isolated muscle can be described by the equation.

$$\frac{dF}{dl} = K P + C$$

Since $\frac{dP}{dt} = \frac{dF/dt}{a}$ as shown previously

equation 3 becomes:

$$\frac{dl}{dt} = \frac{\frac{dP}{dt}\ a}{KP + C} \qquad \text{equation 4.}$$

If there is no change in heart size: equation 4 approximates to:

$$Vce = \frac{dP/dt}{KP}$$

where Vce is the velocity of CE shortening.

Covell et al (1966) produced force-velocity curves in the intact dog heart by plotting a measure of Vce similar to that derived above against myocardial wall tension. Wall tension was calculated from the formula $T = \frac{Pri}{2h}$ where P = transmural pressure, ri = internal radius of the ventricle and h = wall thickness. The relative sensitivity of the force-velocity relationship was compared with that of the ventricular function curve by exerting small inotropic interventions. Noradrenaline infusion always increased Po and maximum Vce (obtained by extrapolation of the curve to zero load) and $\frac{dP}{dt}$ max.

In three out of seven experiments these effects were accompanied by an increase in SW measured at constant LVEDP and MAP. Similarly an increase in HR produced an increase in Vce although Po was increased only very slightly and SW was unchanged. The inconsistency of the effect of noradrenaline and the lack of effect of a change in HR on SW led the authors to conclude that the ventricular function curve was unable to assess alterations in the contractile state of the heart which were reflected primarily by a change in the velocity of contraction, and that the ventricular function curve was also less sensitive than the force-velocity relation in detecting changes in the force of contraction.

In some studies, difficulties with extrapolation to zero load to obtain maximum Vce have been avoided by selecting certain points on the curve relating $\frac{dP}{dt}\Big|P$ versus tension or pressure (Mirsky 1971). Variations in experimental approach and the assignment of different terms to these indices has led to confusion but basically the two most widely used are max $(\frac{dP}{dt}\Big|P)$ and $\frac{dp}{dt}\max\Big|P$, where P is either total or developed pressure in the ventricle; sometimes the factor K is also introduced. These derivatives of $\frac{dP}{dt}$ max are being widely used in clinical situations to assess myocardial contractility, but as Noble (1972) has pointed out, none of these indices are valid under all circumstances and in view of the assumptions which must be made in their measurement, the accuracy of these derivatives in assessing a change in maximum velocity of the CE is highly questionable.

In view of the theoretical and practical problems involved in the measurement in the intact heart of Vmax and the other derived indices, it is doubtful if these indices offer much advantage over the basic measurement of $\frac{dP}{dt}$ max, from which they are derived. For this reason, $\frac{dP}{dt}$ max is used most often as an index of contractility. Furnival et al (1970) emphasised the usefulness of $\frac{dP}{dt}$ max as a sensitive and quantitative index of inotropic changes in the left ventricle. In their experiments a right heart bypass preparation was established in open chest dogs in which it was possible to hold constant LVEDP by varying the output of the bypass pump and MAP by adjusting a screw clamp on the descending aorta. Isoprenaline was infused and its effect on free HR was observed. Changes in $\frac{dP}{dt}$ max were then measured, during the infusion at constant HR, MAP & LVEDP. The changes in $\frac{dP}{dt}$ max were compared with the changes in free HR, which were used as an index of the changes in catecholamine concentration at the SA node. It was shown that changes in $\frac{dP}{dt}$ max were proportional to the changes in HR, which followed stepwise increments in the rate of infusion of isoprenaline. The authors concluded that the changes in $\frac{dP}{dt}$ max were also proportional to the concomitant inotropic changes, and that $\frac{dP}{dt}$ max was thus a quantitative index of inotropic changes. $\frac{dP}{dt}$ max was also compared with SW at constant LVEDP, during the infusion of isoprenaline. Under conditions of constant HR & MAP they found that changes in SW were not consistant with the changes in $\frac{dP}{dt}$ max. SW was, therefore, said to be an unreliable guide to changes in

myocardial contractility.

Furnival et al stressed the conclusion that $\frac{dP}{dt}$ max is the best quantitative index of the action of an inotropic drug on myocardial performance. Implicit in this statement, is the assumption that only one mechanism is involved in controlling contractility, namely the rate of development of tension. Although this is the mechanism by which catecholamines produce their inotropic effect on the heart, it is not the sole mechanism by which contractility can be increased. The mechanical properties of muscle deny the general application of a single index of myocardial contractility since three fundamental relationships provide a complete description of the contractile component;

1. The length-tension curve of active muscle (which is the basis of Starling's Law).
2. The force-velocity curve.
3. The time course of the contraction.

Four variables, the tension in the muscle, the velocity of shortening, the muscle length, and time after excitation, are involved all of which may change simultaneously during a contraction. It has been proposed that a change in myocardial contractility can be defined as a change in the performance of the heart which results from a change in the relationship among these four variables during a contraction. (Blinks and Koch-Weser 1963). This definition does not provide any simple way

of measuring changes in contractility: but it does provide an objective criterion for deciding whether a given change should or should not be regarded as constituting a change in myocardial contractility. Under this definition, only the effects of changes in the fundamental properties of the CE are considered to represent changes in contractility, while changes in performance arising solely from conditions outside the heart are not. For example, a change in the amount of work done in the contraction caused by a change in preload (LVEDP) or in the resistance to ejection (MAP) does not reflect a change in contractility. The relations among the four variables during a contraction can be influenced in various ways, and a thorough characterisation of a change in myocardial contractility requires a full description of the changes.

An increase in myocardial contractility will be manifested by an increase in the strength of contraction. Depending upon the nature of the experiment, this increase in the strength of contraction will appear either as an increase in the peak tension developed or an increase in the amount of shortening in a contraction or both. There are two basic mechanisms by which the strength of contraction can be modified in heart muscle, under conditions of constant initial muscle length and resistance to shortening.

1. A change in the degree of activation of the muscle: which is reflected by a change in the force-velocity relationship of the muscle. In the intact heart, this effect is reflected by

an increase in the rate of development of pressure in the ventricle $\frac{dP}{dt}$. An increase in the degree of activation will normally cause an increase in the peak tension developed unless it is offset by a big decrease in the duration of contraction.

2. A change in the duration of contraction. Under controlled conditions a change in the time to peak tension of an isometric contraction, or a change in the total duration of contraction can be regarded as signifying a change in the duration of activity of the CE. By increasing the time available for tension to develop or for shortening to occur, an increase in the duration of contraction may produce a positive inotropic effect.

It is a dangerous oversimplification to describe a given change in the performance of the heart only as an increase or a decrease in contractility. The current emphasis of looking for a single index of contractility may hamper progress in defining inotropic changes in terms of fundamental processes and ultimately in terms of the underlying cellular and biochemical mechanisms. Attempts are being made in isolated muscle studies to relate inotropic effects more closely to their cellular mode of action and the first step towards this has been the classification of inotropic drugs according to their effects on the time course of the isometric contraction as well as their effect on the rate of development of tension and peak tension (Reiter 1972). Although physiological and pharmacological interventions such as

increased HR, catecholamines and digitales glycosides produce their inotropic effect by increasing the degree of activation there has been an increasing number of reports from isolated muscle studies about drugs which induce a positive inotropic effect by prolongation of the contraction. In the intact heart, changes in the duration of contraction would not be expected to induce changes in $\frac{dP}{dt}$ max: although they might be reflected by changes in SW. In view of this and also of the increasing trend for clinicians to use changes in $\frac{dP}{dt}$ max as an index of contractility although such changes may be unrelated to changes in output of the heart, we decided to study the action of certain drugs and physiological interventions on various indices of contractility to show whether or not these drugs could be classified into groups by their differential action on the parameters studied. Most of the drugs studied were those which had been shown in isolated muscle studies to produce an effect on the duration of contraction. As a change in the duration of the contraction is associated with a change in electrical activity of the myocardium, a brief description of the cardiac action potential seems appropriate.

In contrast to the very short duration of the action potential in nerve and skeletal muscle the cardiac action potential can be 100-500 milliseconds long, depending on the type of heart muscle. Although the electrical properties of cardiac muscle are similar to those of nerve and skeletal muscle in that the action potential is a result of a sequence of changes

in membrane ionic permeabilities, with different ions carrying charges into and out of the cells, the details of the ionic currents in cardiac muscle are not fully understood. This is partly a result of the practical difficulties involved in applying the voltage clamp technique to cardiac muscle and partly because of the greater complexity of the repolarisation process in cardiac muscle. Upon depolarisation to threshold the early inward current is carried by sodium ions, flowing down their electrochemical gradient as a result of a large increase in membrane permeability to sodium ions which arises from the depolarisation. These changes can be described by the same general formulation devised by Hodgkin and Huxley for squid axon. The magnitude of this current is a sigmoidal function of the resting membrane potential value. Reuter (1968) claims that the calcium ions which enter the muscle fibre upon depolarisation also play a role as charge carriers, but the existance and the relative importance of this current is controversial. The early inward current of sodium ions is inactivated at two rates, most of it being inactivated within 10 ms, but there is evidence that a component of the sodium inactivation requires about 100 ms to occur. This slow inactivation contributes to the maintenance of depolarisation during the plateau of the action potential. The potassium currents responsible for repolarisation are quite complex. Noble and Tsien (1969) have postulated the existance of four separate channels. Upon depolarisation to threshold, there is a substantial fall in potassium conductance which helps to reinforce

the depolarising inward sodium current and which also helps to maintain depolarisation during the plateau. Two additional potassium channels, ix_1 and ix_2, are postulated. These are not pure potassium channels as other ions have been found to pass through them. The channel ix_1 is activated in the range of potentials at which the plateau occurs and this time dependant increase in potassium current may be the important outward current in repolarisation from the plateau to the resting membrane potential. This potassium current is variable and has been shown to depend on action potential duration. Ix_2 is too slow to be involved in the normal action potential. The fourth potassium channel is associated with the decreased conductance which occurs after repolarisation and which constitutes the basis of the pacemaker potential.

The multiplicity of currents involved in the cardiac action potential is important not only because of their implications regarding cardiac excitability, but because of the correlation of ionic movements with the contractile activity of the heart. The influx of sodium ions which occurs upon depolarisation of the cardiac cell membrane is accompanied by an influx of calcium (Ca^{++}) ions. The Ca^{++} influx is crucial to the initiation and maintainnce of the contraction. As the concentration of free calcium in the cell exceeds 10^{-7}M, Ca^{++} binds to the protein troponin and by an unknown mechanism, removes the troponin inhibition of bridge formation between actin and myosin. Upon repolarisation Ca^{++} is removed from the myofilaments into

the sarcoplasmic reticulum by an active transport mechanism until the concentration is below the activation threshold concentration of 10^{-7}M, and the muscle relaxes. The duration of the action potential is largely determined by the duration of the plateau after depolarisation. The duration of the plateau, as described previously is determined by the relative ionic conductances of potassium (gk) and sodium, and it is particularly sensitive to gk. A reduced gk will prolong the duration of the plateau, thereby prolonging the influx of Ca^{++} and the duration of the contraction.

The influence of the duration of the action potential on the strength of the contraction is seen particularly in studies on the staircase phenomenon. In frog ventricle, Niedergerke (1956) showed that repetitive stimulation of a previously resting heart produced a progressive increase in contractile tension. But, when this experiment was repeated in a Ca^{++} rich solution, the first contraction was the largest and successive contractions diminished in height. This negative staircase was associated with loss of the plateau and reduction in the duration of the action potential and in time to peak tension as stimulation proceeded. Niedergerke concluded that there were two opposing processes influencing contractility in the staircase phenomenon, one facilitatory and the other inhibitory. Under the experimental condition of a high Ca^{++} concentration, in which the strength of contraction was maximal, the inhibitory effect of a shortening in the duration of contraction was observed.

Also supporting a direct relation between action potential and tension, Kavaler (1959) extended the duration of the action potential in sheep ventricular muscle strips to two seconds, by sustaining the plateau potential with an applied current. He observed that tension was maintained throughout the duration of the plateau and relaxation occured only when the muscle fibres were allowed to repolarise. These observations were extended by Antoni et al (1969), who compared the responses of frog and mammalian myocardium to changes in the action potential duration, induced by constant current pulses. Prolongation of the contraction produced an increase in peak tension of the corresponding contraction only by increasing the time to peak tension. The amplitude of the following contractions, however, initiated by normal action potentials were still increased and were due to an increase in the rate of rise of tension. Five to seven beats were necessary for the development of a steady state. In frog myocardium, however, the contractile response to alterations of the duration of the action potential were limited to the corresponding contraction. The effects of changes in duration of a single action potential on subsequent beats in mammalian myocardium was confirmed by Wood et al (1969).

In order to explain their results Antonii and Wood both postulated the existance of an intracellular store of Ca^{++} ions associated with the transverse tubules and the sarcoplasmic reticulum. The initial effect of a change in action potential duration on the corresponding contraction is postulated to result

from a change in the amount of Ca^{++} entering the cell during that action potential. The increased Ca^{++} influx, however, also increases the size of the intracellular stores of Ca^{++} available for release by subsequent action potentials. Frog myocardium, in which the transverse tubule -sarcoplasmic reticulum is very poorly developed does not show the increased $\frac{dT}{dt}$ in subsequent beats. Many experiments using radio active Ca^{++} to measure Ca^{++} influx during depolarisation have shown that only a small fraction of the activator Ca^{++} originates directly from the extracellular space (Langer 1973). These and other studies on the binding of Ca^{++} ions by the sarcoplasmic reticulum support the hypothesis of an intracellular store of Ca^{++} ions, in mammalian hearts.

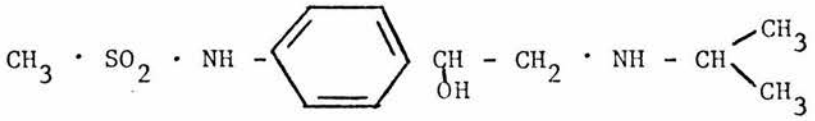
The duration of contraction is determined by the balance between the time course of Ca^{++} influx and the activity of the Ca^{++} sequestration system. An increase in frequency of stimulation and catecholamines usually produce a positive inotropic effect because the increased rate of arrival of Ca^{++} at the troponin sites more than compensates for the negative inotropic effect of a decreased duration. Interventions which produce a positive inotropic effect, based on prolongation of the contraction have only comparatively recently been investigated. The action of caffeine in this respect has been most closely studied. Degubareff and Sleator (1965) using guinea pig atria showed that the positive inotropic effect of caffeine was associated with an increase in the duration of the action potential and in the duration of the contraction, this effect being dependant on Ca^{++}

concentration. These results were confirmed by Gibb (1967) Caffeine was shown to prevent the effect of adrenaline on the relaxation phase of the contraction such that adrenaline in the presence of caffeine, no longer abbreviated the contraction. The effects of caffeine on force development in myocardium are consistant with a dual action of caffeine on excitation-contraction coupling as proposed by Blinks et al (1972). In their experiments on kitten papillary muscle, caffeine not only increased the duration of contraction but also increased the maximal rate of tension development $(\frac{dT}{dt}$ max). Procaine antagonised the caffeine induced prolongation of duration of tension development, but did not affect the initial rapid increase in $\frac{dT}{dt}$ max and peak tension. It was, therefore, proposed that caffeine increased the duration of contraction by inhibiting Ca^{++} sequestration by the sarcoplasmic reticulum, an action which is procaine sensitive: and also is capable of inducing Ca^{++} release, which is procaine insensitive. These concepts are supported by work done on isolated frog and rabbit skeletal sarcoplasmic reticulum preparations. (Weber and Herz 1968). It was noted that caffeine produced a release of Ca^{++} from reticulum preparations loaded with Ca^{++} and it also reduced the rate of Ca^{++} uptake by the sarcoplasmic reticulum.

High concentrations oftyramine, cevadine, veratridine, sotalol and bretylium are among the other agents recently reported as having a slowing action on the relaxation phase of contraction paralled by a characteristic prolongation of the

action potential. Although this action has only been reported in isolated muscle, there has been some controversy over the positive inotropic effect of sotalol and bretylium in vivo. We decided to investigate the positive inotropic effect of sotalol and bretylium in vivo to determine whether this action maybe attributable in part to their effects on the duration of the contraction.

Sotalol, 4'- (1- hydroxy-2-(isopropylamino)ethyl) methane sulphonanilide Hcl: formula



is a beta adrenergic blocking drug. Three actions of sotalol on myocardial contractility have been reported.

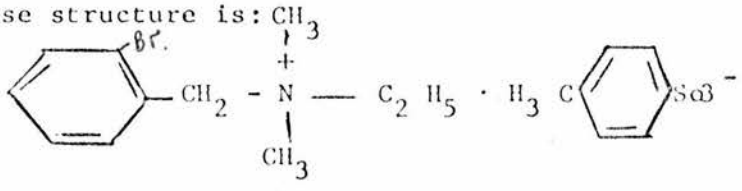
1. an indirect depression caused by blockade of the effect of circulating catecholamines on the heart.
2. a direct negative inotropic effect.
3. a positive inotropic effect, produced by prolongation of the contraction.

Blinks (1967) observed that sotalol antagonised the inotropic and chronotropic effect of catecholamines and in very high concentrations also produced a direct depression of the strength and frequency of contraction of the heart. Superimposed on this depression of contractility, sotalol had a stimulant action on the strength of contraction in cat ventricular muscle

but not atrial muscle. This positive inotropic effect was not mediated via beta receptors in the heart since it was not prevented by propranalol. (Kaumann & Blinks 1967). The developed twitch tension was greatest in a concentration of 10^{-4}M sotalol. At this concentration, sotalol slowed the terminal phase of relaxation and a small amount of active tension, which the authors named an aftercontraction, persisted after each contraction. The aftercontractions were associated with a marked prolongation of the action potential (Kaumann and Olson 1968) With long exposure to the drug, the duration of the action potential and the twitch plus aftercontraction could be increased three to five fold from the control. The effect of sotalol on the duration of contraction was also investigated by Parmley et al (1972). Comparing sotalol with propranalol on cat papillary muscle contracting isometrically, they showed that sotalol, in a concentration of 10^{-4} M to 10^{-3}M, caused an increase in contractile force accompanied by a prolongation of the contraction: whereas propranalol at doses greater than 10^{-6}M had a negative inotropic effect, and caused shortening of the duration of the contraction. At a dose of 10^{-2}M, however, sotalol caused a marked depression of contractile force. The positive inotropic effect of sotalol was unchanged in reserpine pretreated muscles. A detailed examination of the effect of sotalol on the electrophysiological properties of cardiac tissue has been carried out by Strauss et al (1970), and Singh and Vaughan Williams (1970). Strauss et al showed that sotalol, in doses up to 5.10^{-4}M,

had no action on the resting membrane potential, on overshoot amplitude and on the maximum rate of rise of the action potential in dog purkinje and ventricular muscle fibres. Under conditions of constant cycle length, sotalol in concentrations between 10^{-5} and 10^{-3}M, prolonged the duration of the action potential as a result of lengthening of the plateau of the action potential and particularly the terminal phase of repolarisation. These results were confirmed by Singh and Vaughan Williams in cat papillary muscle. They also reported that sotalol prolonged the Q-Tc interval with no change in the P-R interval of the E.C.G. recorded in anaesthetised guinea pig. This effect was, however, associated with a slowing of the spontaneous heart rate. In the intact heart, conflicting results have been reported. Hoffman and Grupp (1969) noted that sotalol reduced ventricular contractile force in both normal and reserpinised anaesthetised dogs, indicating a direct negative inotropic effect. Puri and Bing (1969), however, concluded that sotalol had no direct myocardial depressant action and that the fall in $\frac{dP}{dt}$ max associated with a fall in HR, was due to blockade of the action of circulating catecholamines. Systolic time was prolonged and therefore stroke output and stroke work remained unchanged. The lack of a direct depressant action of sotalol was also noted in man (Svedmyr et al 1970).

Bretylium tosylate is a quatenary benzyl ammonium salt: whose structure is:



The ability of bretylium to increase myocarial contractility was first described by Boura & Green (1959) who introduced the drug as a hypotensive agent. The basis for bretylium's hypotensive action was its ability to block the release of noradrenaline from adrenergic nerve endings. It's blocking action was preceded by an initial release of noradrenaline. Subsequent studies confirmed a positive inotropic effect of bretylium but disagreed about its importance and mechanism. Gilmore & Siegel (1962) reported that, in the dog, bretylium administration like cardiac sympathetic nerve stimulation produced an increase in myocardial contractility which was accompanied by an increase in the coronary venous catecholamine levels. They concluded, therefore, that the myocardial effects of bretylium were entirely indirect and were due to the release of endogenous catecholamines. Gaffney, however, was able to show a small positive inotropic effect of bretylium on a dog heart-lung preparation depleted of catecholamines by prior administration of reserpine and in the complete cardiac denervated open chest dog. (Gaffney 1961: Gaffney et al 1962). The positive chronotropic effect of bretylium in the normal heart, which is associated with release of noradrenaline, was reversed to a negative chronotropic effect in the reserpinised heart. Gaffney concluded that in addition to its indirect actions, bretylium had some direct positive inotropic effect on heart muscle. This conclusion was also drawn by Amsterdam et al (1970) who reported a positive inotropic effect of bretylium on cat papillary muscle in the presence of beta adrenergic blockade with propranalol. As the

literature contained conflicting data on the question of whether the inotropic effect of bretylium was due to catecholamine release or a direct effect on the myocardium, Markis & Koch-Weser (1971) re-examined the action of bretylium on isolated mammalian myocardium. The positive inotropic effect of bretylium on kitten papillary muscle, contracting isometrically, was accompanied by an increase in the rate of development of tension with a very slight decrease in the time to peak tension and no change in the time to 90% relaxation. Noradrenaline, however, shortens relaxation time and the duration of contraction is decreased. On reserpine pretreated muscle, the positive inotropic effect was much reduced but relaxation was slowed and consequently the duration of contraction was increased. Bretylium was thus shown to have a direct effect to slow relaxation. The failure of bretylium to alter the relaxation time of normal myocardium reflects the cancelling out of its direct effect on this parameter of contraction and the opposing effect of released noradrenaline. Although bretylium increases myocardial contractile force mainly by the release of endogenous noradrenaline, the net inotropic effect must reflect the interaction of this indirect action with its direct action tending to prolong the duration of the contraction.

METHODS

1. Control of anaesthesia, arterial blood gas tensions, acid-base balance and temperature.

Mongrel dogs and cats were used as the experimental animal species. Dogs wighing between 5 and 10 kg. were anaesthetized by an intravenous injection of pentobarbitone sodium 30 mg/kg. Cats weighing between 3.0 and 5.3 kg were anaesthetized by an intraperitoneal injection of pentobarbitone sodium 40 mg/kg. As changes in the depth of anaesthesia alter the parameters which we were studying, a constant level of anaesthesia was subsequently maintained by a continuous infusion of pentobarbitone sodium, 6 mg/ml, into a femoral vein from a Palmer slow infusion pump. The rate required was of the order 10-30 mls/hour for a dog, depending upon the animals weight and the level of anaesthesia, as indicated by muscle tone and the state of its corneal reflex.

The trachaea was cannulated and the animal ventilated with 100% oxygen using a variable stroke Starling 'ideal' pump, running at twenty strokes per minute. Changes in blood gas tension and pH can directly affect the rhythm and the contractile state of the heart. It was, therefore, necessary to maintain these parameters within strict limits during the experiment. Throughout the experiment, samples of arterial blood were withdrawm at regular intervals and pH, Pco_2 and Po_2 measured using a Radiometer BMS3 system.

Arterial Pco_2 was kept as near possible to 28 torr in the cat and 40 torr in the dog, by adjusting the stroke of the respiratory pump. Plasma bicarbonate concentration was determined using the nomogram of Siggaard-Anderson (1963) and corrected to 18 mM in the cat and 25mM in the dog, when a base deficit had developed, by an intravenous injection of an appropriate volume of 1M sodium bicarbonate solution.

Rectal temperature was recorded from a thermistor probe and was held near to 37°C by a heating pad below the animal and also when necessary an infra-red lamp above the animal.

II. Operative procedure.

Incisions were made and bleeding points coagulated using a Gimber veterinary diathermy unit. Aortic blood pressure was recorded from a catheter inserted through the right common carotid artery. Positive expiratory pressure ventilation was applied and the chest opened through a mid-sternal split. The heart was denervated by cutting the vagi and crushing the sympathetic ansae. The pericardium was opened and when the connective and fatty tissue around the aorta and the pulmonary artery had been cleared away, an electromagnetic flow probe (Statham Q Series) was placed around the ascending aorta. A bipolar platinum electrode (20 mm long) was inserted into the

right auricular appendage and the heart was paced, using 5 V pulses of 5 ms duration at a frequency just faster than the spontaneous rate. A purse string was sutured around the apex of the left ventricle. The animal was heparinized (1000 u /kg) and a metal cannula (internal diameter 2.2 mm: length25mm) with side holes was inserted through the apical dimple of the left ventricle. Consolidated electrodynamic L223 transducers were attached to the left ventricular and aortic pressure cannulae and the signals amplified.

III Parameters measured

The dynamic response of the ventricular pressure recording system was examined in order to be certain that the pressure pulse contour which we recorded was an accurate reproduction of the pressure changes in the ventricle. This analysis was not necessary for the aortic pressure recording system as we were mainly interested in the recording of mean arterial pressure. It is assumed that any pressure function of time existing in the circulation is a periodic function and as such constitutes what is known as phasic oscillation. The simplest form of phasic oscillation is the sine wave. By a Fourier analysis, any complex phasic oscillation can be split down into a series of sine waves, each being an arithmetic progression of frequencies. The general equation for a Fourier series is

$$f(t) = A \sin (wt + a) + B \sin (2wt + b)... + Z \sin (Nwt + n)$$

each term being known as a harmonic, From a Fourier analysis, the number ofharmonics of significant amplitude necessary to inscribe a pressure pulse indistinguishable from the original can be calculated. Such analysis of typical ventricular pressure pulses have indicated that it is necessary to record accurately to the tenth harmonic of the pulse wave. Hansen (1950) has shown, however, that in most cases the pressure pulse can be reproduced with 95% accuracy by the first six harmonics.

In order to measure the frequency response of the ventricular pressure recording system a square wave pressure change was applied to the system by bursting an inflated balloon attached to the transducer system. By applying the formulae derived by Hansen and Warburg (1950) to the series of oscillations obtained upon the square wave pressure change, the frequency response of the system was obtained as follows:-

The damping factor B, was calculated from the equation

$$B = \frac{\lambda}{\sqrt{4\pi^2 + \lambda^2}} \qquad \text{equation 1.}$$

where λ is the difference between the natural logarithms of consecutive overshoots on the same side of equilibrium.
The undamped natural frequncy is given by

$$fo = \frac{\sqrt{4\pi^2 + \lambda^2}}{2\pi T_D} \qquad \text{equation 2.}$$

where f is frequency is cycles/sec.

T_D is the period of one complete oscillation.

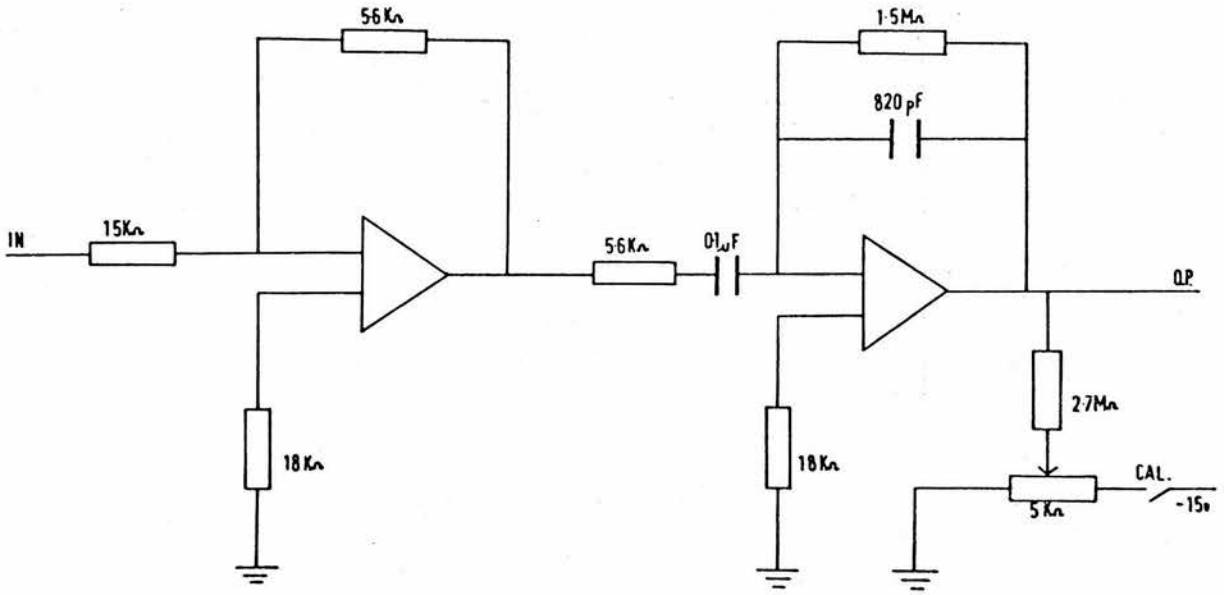
From the amplitude-frequency curves for oscillation systems with different degrees of damping (Hansen & Warburg 1950) the value of Y_R (which is the ratio of the resonant frequency to the undamped natural frequency) at which the amplitude becomes either greater than 1.05 or less that 0.95 is found on the line with the value of B obtained from equation 1. This value of Y_R, multiplied by Fo obtained from equation 2, represents the highest frequency which the system will reproduce with better than 5% accuracy. Apart from the possible amplitude distortion there is the possibility of phase distortion. The phase difference angle is dependant upon the relative frequency, Y, and the degree of damping. From the graph relating relative phase difference time and the influencing frequency for different degrees of damping (Hansen and Warburg 1950), it can be seen that the relative phase distortion is less the lower the degree of damping is. Our system had a damping factor B = 0.2 and so phase distortion was practically negligible. The frequency response of the ventricular pressure recording system, obtained by this method, was flat to 24 Hz. Since in our experiments the most rapid fundamental frequency encountered was of the order of 4 Hz and Hansen states that a pressure pulse can be reproduced with 95% accuracy by the first six harmonics, the frequency response of this system is sufficient for accurate reproduction of the ventricular pressure pulse.

The transducers were calibrated in a stepwise manner using a mercury manometer. During each experiment, frequent checks against a standard pressure, atmospheric pressure, allowed drift to be recognised. The transducer connected to the ventricular cannula was calibrated with the base of the heart as the reference point.

Aortic pressure, left ventricular pressure and aortic flow were recorded both on magnetic tape (Bell and Howell VR 3200) and on light sensitive paper (Honeywell Visicorder). The transducer amplifiers were also connected to the following parallel analogue circuits.

I) An analogue differentiating circuit to derive $LV\frac{dP}{dt}$.
The circuit for derivation of $LV \frac{dP}{dt}$ is shown in Figure 1. In response to sine wave changes in the input voltage, the output of the differentiator showed a 90^{o} phase shift and there was a linear relationship between the amplitude of the output and the frequency of the input signal from 1 to 128HZ. The recorded output was calibrated using a variable slope waveform generator to provide an input to the differentiator. The output of the generator was expressed in volts/sec. Since the output amplitude of the left ventricular amplifier was known to be 2.3 volts output for 100 mm Hg, by comparing the differentiated ramp amplitude and the $\frac{dP}{dt}$ left ventricular amplitude a calibration value for the differentiator was obtained. Frequent checks were made to ensure that the differentiator remained calibrated at 5000 mm Hg s^{-1}.

Fig. 1 CIRCUIT DIAGRAM FOR DIFFERENTIATOR



II) An EAL 380 analog computor to derive LVEDP and external SW beat by beat.

The external SW of the left ventricle is usually calculated as the sum of two components, one the work done against pressure and the other, the work done in imparting velocity to the aortic stream. The work done against pressure is given by the formula

$$W = \int_{t}^{t'} PdV$$

where t-t' marks the beginning and end of systole: P is the pressure of each increment of volume (dV) ejected.
The work done in accelerating the blood is:

$$\int_{t}^{t'} \frac{v^2 \quad dM}{2g}$$

where v is the velocity of each increment of weight (dM) ejected, g is the gravity constant. Remington and Hamilton (1947) calculated the velocity component of the external SW in dogs and showed that at rest, it is of the order of 2% of the total external work. As this function can be considered to be insignificant in dogs at rest, it is usually omitted in the calculation of external SW.

We calculated external SW according to the equation -

$$SW = \int_{t}^{t'} F\ (LVP - LVEDP)\ dt$$

where F is the instantaneous aortic flow, LVP is the instantaneous left ventricular pressure and $t' - t$ is the systolic phase of a cardiac cycle. Most people calculate external SW, by making the assumption that the whole of the stroke volume is ejected against a constant pressure. SW can then be calculated using the mean values of aortic pressure and stroke volume if the following assumptions are made. The work done is equal to the force developed (F) multiplied by the distance it moves (D).

i.e. $W = F \times D$ equation 3.

Since $F = P \times A$ and $D = \frac{V}{A}$

Where P is the pressure increase imparted to the blood by the heart, V is the volume of blood ejected, and A is the cross sectional area of the aorta. equation 3 becomes:-

$$W = PA \times \frac{V}{A}$$
$$= PV$$

The calculation of external SW is therefore often approximated, according to the formula - $SW = (MAP - LVEDP)\ SV$

The errors involved in this approximation are:

(1) MAP is less than the pressure during ejection, so that SW is underestimated. This underestimate must increase if ejection time becomes a smaller fraction of a cardiac cycle. This occurs if catecholamines are infused at a constant paced heart rate.

(2) Pressure and flow are out of phase such that flow is still increasing when pressure is falling. Using a constant value of pressure in the approximation will tend to overestimate SW since because of the phase difference, pressure is proportionally smaller for each value of flow.

In some experiments we also calculated SW, using the mean

values of pressure and volume and compared these values with those obtained using the integrated data. Both values of SW were calibrated in milli Joules (mJ), by using the appropriate conversion factor.

The analogue computation necessary for the calculation of SW and the reading of LVEDP is shown in Figure 2. After the inputs have been scaled, LVEDP was obtained from the diastolic phase of the LVP curve. The LVP record and the pulse responsible for the reading of LVEDP were displayed on an oscilloscope so that the point in the cardiac cycle at which LVEDP was being read, could be checked and corrected, if necessary. LVEDP is continuously substracted from the following LVP. LVP - LVEDP is then multiplied by the instantaneous aortic flow, and the area under this curve is integrated to provide a value of external SW.

Parralel logic was used to set the limits of each cardiac cycle in the computation of SW, to read LVEDP, and to store the derived parameters during each cardiac cycle. A comparator was used to detect the point in the left ventricular pressure curve, at which pressure begins to rise. When the LVP is less than the reference voltage: i.e. when the heart is in diastole, a logic '1' level is fed into and gate F. If the other input into and gate F is also logic '1', the track/store amplifiers for LVEDP and SW will begin to track. When the LVP begins to rise above the reference voltage, the output of the comparator changes to logic 'o', causing the track/store amplifiers to store the values of LVEDP and SW they had at the instant of switching.

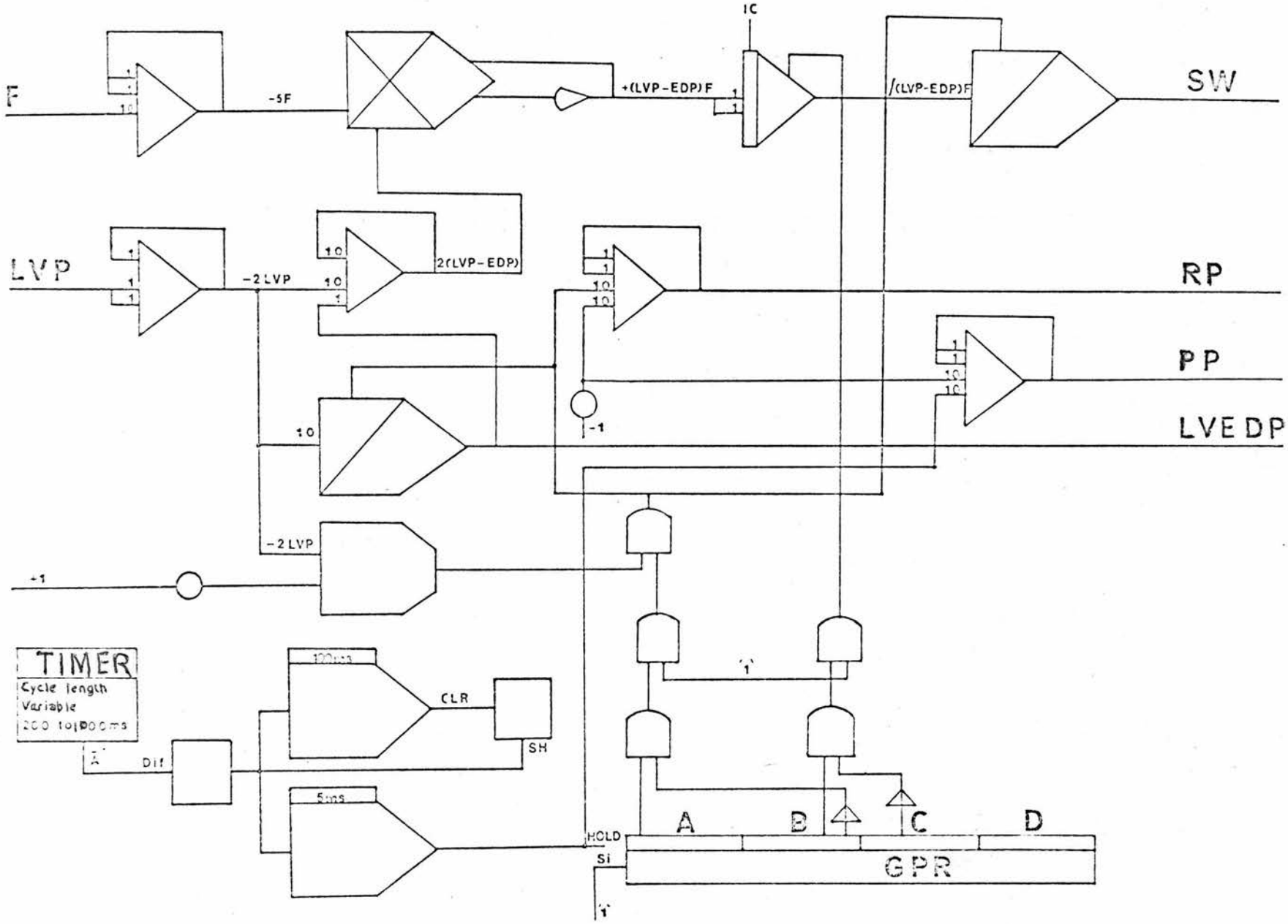


Figure 2. The analogue computation for the calculation of SW and LVEDP. PP and RP are the pacemaker and reading pulses respectively.

This occurs if there is a premature systole, in which case the T/S continues to read the LVEDP for the previous beat. A source of pulses is used to control the conditions of the other and gates in the logic net. These pulses, of cycle length which is variable between 200 and1000 ms, are used to provide the pulses which drive the pacemaker in the right atrium. The animal's HR can, therefore, be set by setting the cycle length of the pulses. The duration of the pacemaker pulses (PP) was controlled by a monostable, which was set at 5ms. For the duration of the pacemaker pulse, the integrator is maintained in a hold mode, so that the stimulus artifact which is detected by the flowmeter is not integrated. The source of pacemaker pulses also governs the control of a shift register, such that each bit of the shift register will change state to a logic '1' output, with a very short delay between the changing of each bit. This delay circuit is necessary so that the integrator can be reset by and gate E, a short time after the value of SW has been read. When the output of register A is logic '1' and the normal output of register B is logic 'o' the output of and gate A is logic '1'. The output of and gate C is, therefore, also logic '1'. If the heart is in diastole, as indicated by the logic output of the comparator, the output of and gate F would also be logic '1'. This logic '1' instructs the track/store amplifiers to track the values of SW and LVEDP. The logic '1' output of and gate F is, therefore, the pulse responsible for the reading of LVEDP, this pulse (RP) occuring with a set delay after

the pacemaker pulse. The shift register is cleared after a delay governed by a monostable.

The pacemaker pulses were scaled so that when they were recorded on magnetic tape, they were 1 volt in height above a baseline of 0.5 volts. The three derived signals, $\frac{dP}{dt}$ SW and LVEDP were also recorded on magnetic tape and on light-sensitive paper. LVEDP and MAP were displayed throughout the experiment on galvonometers: SW on a digital voltmeter and the other parameters on a 4 channel oscilloscope. Calibration signals representing known amounts of each parameter measured were also recorded on the tape.

IV Data Analysis

The data recorded on magnetic tape were later analysed using a PDP8 digital computer. The maximum voltage output of the tape is 1.5 volts, but since the analogue to digital converter works within the range 1 to 10 volts, each output on the tape was multiplied five times before being fed into the analogue - digital converter. The data analysis was carried out on a beat to beat basis. Each parameter being measured had, therefore, to have a static value. This had already been accomplished for SW and LVEDP by the track/store amplifiers of the analogue computer. Mean aortic pressure and cardiac output were obtained by passing pulsatile aortic pressure and pulsatile aortic flow signals respectively through averaging circuits before entering

the digital computer. An analogue peak height detector unit was used to obtain positive $\frac{dP}{dt}$ max from the record of $\frac{dP}{dt}$. The maximum leakage from the capacitor through the resistor which occurs before $\frac{dP}{dt}$ max is read is about 2% of the value of $\frac{dP}{dt}$ max. This is a consistant error and it was considered to be negligible in the reading of $\frac{dP}{dt}$ max.

The computer was programmed to recognise a change in voltage in the pacemaker pulse channel, from 2.5 volts to 7.5 volts, (i.e. one pacemaker pulse) as the beginning of each cardiac cycle. Each of the other five channels were then read with a delay of 60us between each reading. The total delay involved in the reading of all the channels was 360 ms a delay of this magnitude would not affect the readings over each beat. The digital computer was calibrated using the calibration signals on the tape and the known values of each calibration signal. The mean and standard error of mean (SEM) of all the parameters were calculated over a sequence of ten beats within each test. Before a print out of the mean values of the parameters was obtained, the data from all ten beats was displayed on a videoscope (Telephonix 401D). If that particular sequence of beats contained erroneous readings or if missed beats had occurred, the data was rejected and another ten beats in the test was analysed.

The reliability of the values obtained from the digital computer was evaluated by frequent spot checks of the measurements

recorded on the UV paper.

In those experiments, in which we wanted to observe an action on free HR, the HR was measured using the ECG. The ECG was obtained through an electrode attached to the ventricular cannula and an indifferent electrode. The R wave of the ECG was used to trigger a ramp generator back to baseline, the amplitude of the record produced being proportional to the R-R interval. The device was calibrated by determining the amplitude equivalent to one second.The free heart rates obtained using this method were only recorded on UV paper. In the experiments, in which we were looking for an effect on the duration of the electrical and mechanical activity, we recorded the ECG at constant HR, MAP and LVEDP during each test on magnetic tape.

Duration Parameters.

The durations of the mechanical and electrical activity in each beat were measured by playing back the pulsatile ventricular pressure, the pulsatile aortic pressure and the ECG records from the tape onto a fast running UV paper. Three measurements were made.

1. The time between the upstroke and the return to diastole of the ventricular pressure curve, which was used as an indication of the duration of the contraction.
2. The time from the onset of rise of the aortic pressure to

the closure of the aortic valve, as indicated by the incisura notch. This measurement was taken as an indication of the duration of systole.

3. The time between the upstroke of the Q wave on the ECG trace to the peak of the T wave.

V. Method of controlling or changing HR, MAP and LVEDP

As stated previously, the HR was controlled by pacing of the right atrium. A change in HR was produced by adjusting the clock of the analogue computer to produce pulses of the desired cycle length.

In the cat MAP was held constant by compression of the descending aorta against a cotton tape which had previously been placed around it. LVEDP was held constant by injection of a dextran solution or removal of blood.

Although this method was satisfactory in the cat, we found that in the dog the compression of the aorta by a screw clip did not provide sensitive enough control over MAP. For this reason a different method of controlling MAP and LVEDP was used in the dog. A compressed air blood pressure compensator was used which consisted of an inverted conical flask, of volume 2 litres, attached to a compressed air supply inlet: a screw clip which was used to regulate the air pressure in the reservoir: and an anaeroid barometer for the measurement of

the pressure in the reservoir. The blood pressure compensator was attached to the animal via both femoral arteries. Care was taken to maintain the resistance of the cannulae and the connective tubing at the lowest possible value, by using the largest bore cannulae possible. If the animals blood pressure rose above the pressure set in the reservoir, blood would move out of the animal down the pressure gradient, until the pressure in the animal and the pressure in the reservoir were equal. Similarly, if the animal's BP fell below that in the reservoir, blood would move in from the reservoir until the pressure gradient was zero. In order to prevent the accumulation of blood in the BP compensator when there was a rise in BP, the compensator was connected through a Watson-Marlow pump to a reservoir. The reservoir was connected to a silicone rubber cannula (3mm id) with side holes, inserted through the left auricular appendage, so that its tip lay in the atrium.

The system was primed with a solution containing dextran (2.5% W/V) and glucose (5% W/V). By transfer of fluid from the venous to the arterial side of the circulation or vice versa both MAP and LVEDP could be maintained constant or varied independantly as desired. A water jacket and a heated glass coil in the reservoir were used to maintain the fluid in the system near 37^{o}C. All the parts of the system which were made of glass were siliconed to prevent the formation of pharmacologically active agents when the blood contacted the glass.

VI Drug administration

Drugs were given after data had been recorded for twenty minutes to ensure that a steady state level had been achieved. Noradrenaline bitartrate was given by continuous intravenous infusion using a Watson-Marlow pump, at a rate within the range 0.05to 0.41 ug/kg/min. The heart was paced at a rate just exceeding that of the expected spontaneous rate induced by noradrenaline. The resting levels of all the parameters being studied were recorded just before the infusion began, the values of HR, MAP and LVEDP being noted. During the infusion of noradrenaline, HR, MAP and LVEDP were held constant at their preinfusion levels and when a steady state had been achieved the parameters were again recorded.

Sotalol, propranalol and bretylium were injected intravenously in doubling cumulative doses. The parameters were recorded before and 5 minutes after the administration of each dose of drug; HR, MAP and LVEDP being held constant when necessary. The haemodynamic responses to each of these drugs were observed following this protocol in normal dogs, in dogs which had been pretreated with reserpine in order to deplete then of catecholamines and in dogs in which blockade of the effects of circulating and released catecholamines on the heart was achieved with practalol 10 mg/kg.

0.5 mg/kg of reserpine was given intramuscularly on each of the two days preceding the experiment, and a third dose was

given intravenously at the start of the experiment. This dosage of reserpine is reported to result in virtually complete depletion of measurable noradrenaline in the heart within 24 hours (Paasonen and Krayer 1958). Tyramine, 60 ug/kg, was given intravenously to test for depletion of endogenous catecholamines in the reserpinized animals. Absence of a change in HR or MAP to this dose of tyramine is claimed to indicate complete depletion (Cooper 1961).

Prostaglandin solutions were infused in the cat and dog into the left ventricular cavity through the indwelling catheter at a rate of 0.5 or 1.0 ml/min for two minutes. Since prostaglandin C is unstable under alkaline conditions, fresh solutions were prepared in 0.9% Nacl solution at ph 5.5, from a 3mg/ml stock solution in methanol, at the start of each experiment. Prostaglandin (PG) C_2 was prepared from PG A_2 using rabbit plasma PGA isomerase bonded to sephatose 4B gel (Jones 1974). Two kinds of tests were performed using prostagladins. In one series (controlled tests) MAP, HR and LVEDP were held constant during the prostaglandin infusion, so that only direct actions on the heart were seen. In the other series (uncontrolled tests) MAP and LVEDP were allowed to change freely under the action of the drug, so that actions on cardiac performance secondary to changes in arterial pressure and venous return were seen. After the first infusion of prostaglandin there was marked tachyphylaxis in the respones to further doses of prostaglandin. We therefore, adjusted the dose of prostaglandin to keep the same order of fall in blood pressure in the uncontrolled

tests. Similar allowance was made for the variation, which was greater than tenfold, in sensitivity to prostaglandins between animals.

VII Expression of results.

The results have been expressed, either as the absolute change or as the percentage change from control valves. Where the results have been pooled, each bar represents the mean $\pm$ 1 standard error of mean (SEM). For each group of experiments the control measurements have been expressed as the mean $\pm$ 1 standard deviation (SD). Regression equations were calculated by the method of least squares fit. Student's 't' test was used to evaluate the significance of a change from the control level, changes being considered significant if $P<0.01$

S E C T I O N 1.

R E S U L T S

Effect of heart rate on $\frac{dP}{dt}$ max, SW and cardiac power.

By adjusting the pacemaker, stepwise increments in heart rate were brought about. The results of five series of tests in five dogs are shown in Figures3&4 and the data tabulated in table A . The mean lowest heart rate was 150 beats/min (SD^{+}22), the mean MAP was 79 mm Hg (SD^{+} 5), and the mean LVEDP was 2.8 mm Hg (SD^{+} 1.5). During each series of tests, MAP and LVEDP were held constant within $^{+}_{-}$ 3 mm Hg and $^{+}_{-}$ 0.3 mm Hg respectively.

In each animal an increase in HR was accompanied by an increase in $\frac{dP}{dt}$ max. The calculated regression equation indicates an increase in $\frac{dP}{dt}$ max of 66 mm Hg S^{-1} for an increase in HR of 10 beats/min (Figure 3). In two out of five experiments SW rose to a maximum value, then fell as HR was further increased. In the other three experiments SW fell as HR was increased. (Figure 4). Mean cardiac power was calculated as the product of HR and SW. It fell as HR was increased in one experiment and in the other four, mean cardiac power rose to a maximum value then fell as HR was further increased (Figure 4).

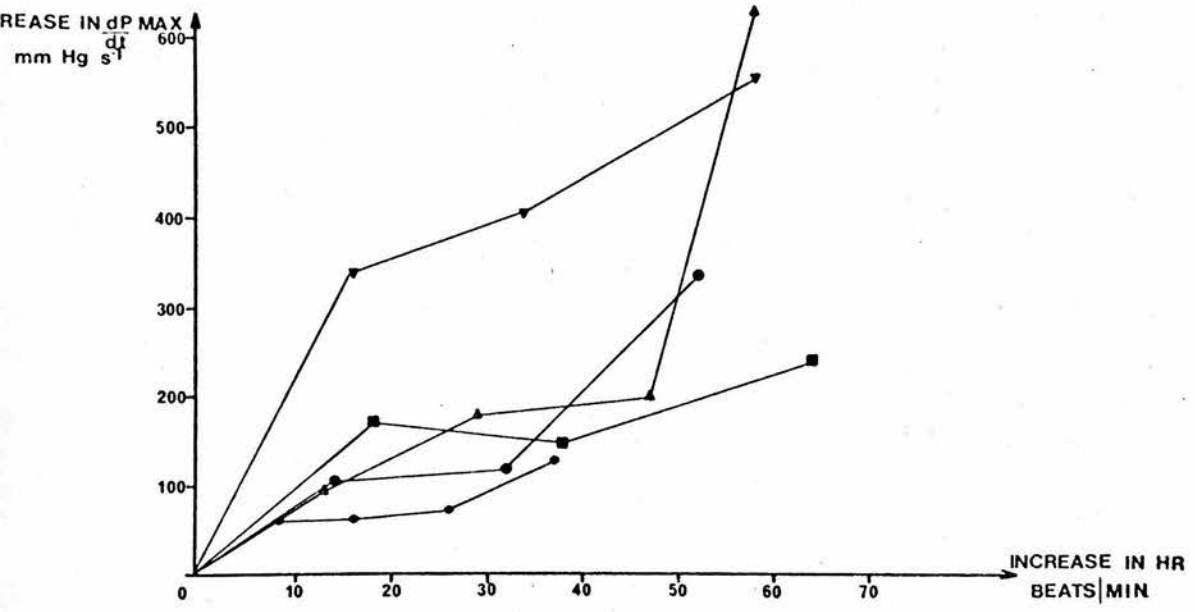


Figure 3. The effect of increasing HR on $\frac{dP}{dt}$ max.

The relationship between the increase in HR and the change in $\frac{dP}{dt}$ max. The mean lowest $\frac{dP}{dt}$ max was 1404 mm Hgs^{-1} (S.D. $\pm$ 153). Each symbol represents the results obtained in one dog. The regression equation is Y = 6.6x +10.

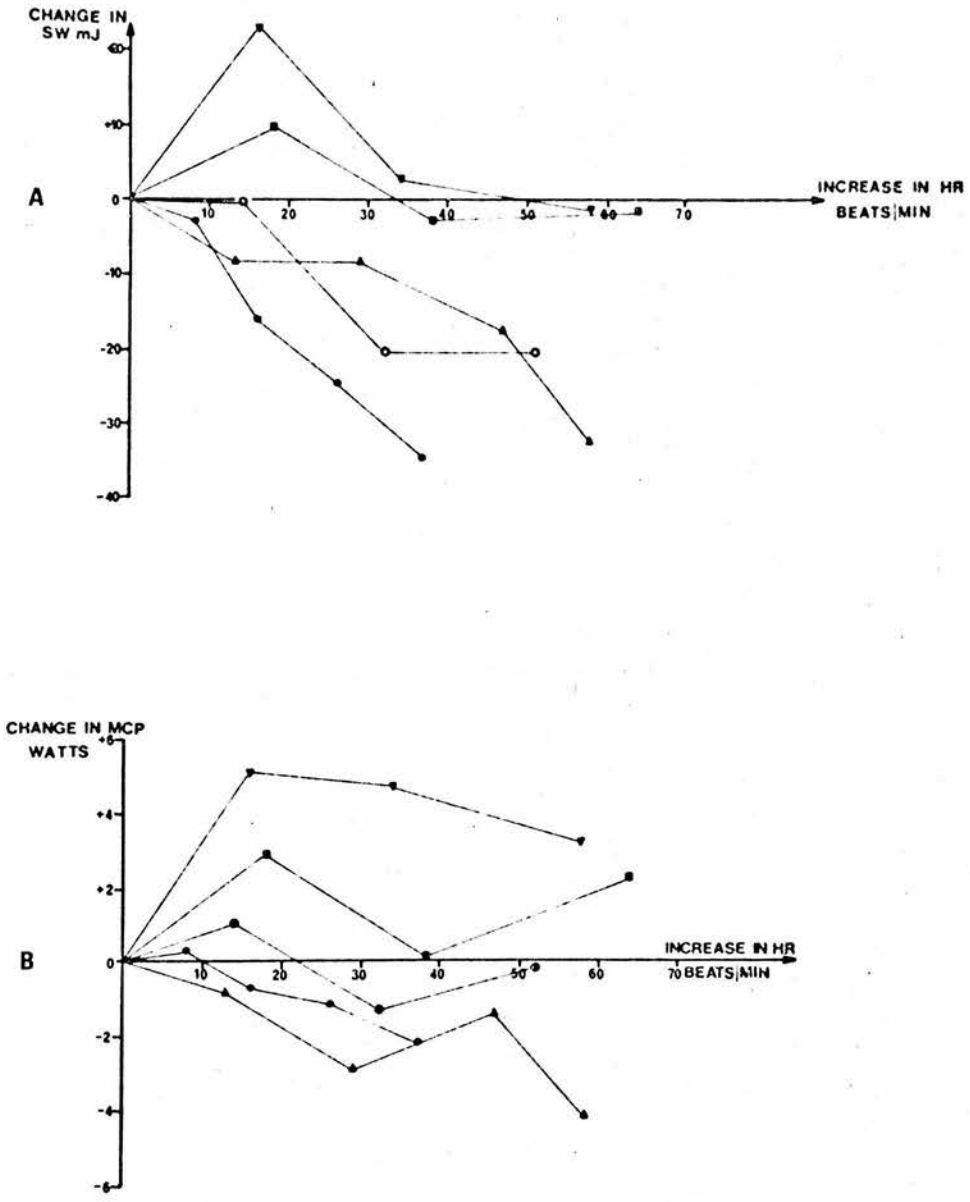


Figure 4. The effect of increasing HR on SW and MCP. Each symbol represents the results obtained in one dog.

A. The relationship between the increase in HR and the change in SW. The mean lowest SW was 60.6 mJ (S.D. ± 18.7).

B. The relationship between the increase in HR and the change in mean cardiac power (MCP) MCP (watts) =HR (beats/~~min~~ sec) x SW (mJ). The mean lowest MCP was $\frac{8.0}{60}$ watts (S.D.± 0.6)

Effect of increased afterload on $\frac{dP}{dt}$ max and SW.

In five dogs MAP was altered by stepwise increments or decrements of the air pressure in the blood pressure compensator. The results are shown in Figure 5 , and the data tabulated in table B .

The mean HR was 153 beats/min (SD$^{\pm}$ 9): the mean LVEDP was 3.2 mm Hg (SD$^{\pm}$ 1) and the mean lowest MAP was 71 mm Hg (SD$^{\pm}$ 16). During each series of observations, HR was held constant and LVEDP was held constant within $^{\pm}$ 0.4 mm Hg.

In each animal an increase in MAP was accompanied by an increase in $\frac{dP}{dt}$ max and in SW. The calculated regression equations indicate that for a rise in MAP of 10 mm Hg, there is an increase in $\frac{dP}{dt}$ max of 86 mm Hgs^{-1} and an increase in SW of 8 mJ.

Effect of increased preload on $\frac{dP}{dt}$ max and SW.

In 6 dogs, LVEDP was increased in a stepwise fashion by infusion of dextran into the left atrium. The results are shown in Figure 6 , and tabulated in table C .

The mean HR was 153 beats/min (SD$^{\pm}$ 5), the mean MAP was 77 mm Hg (SD$^{\pm}$ 6) and the mean lowest LVEDP was 1.1 mm Hg (SD$^{\pm}$ 0.8). During each series of observations, HR was held constant and MAP was held constant within $^{\pm}$ 5 mm Hg.

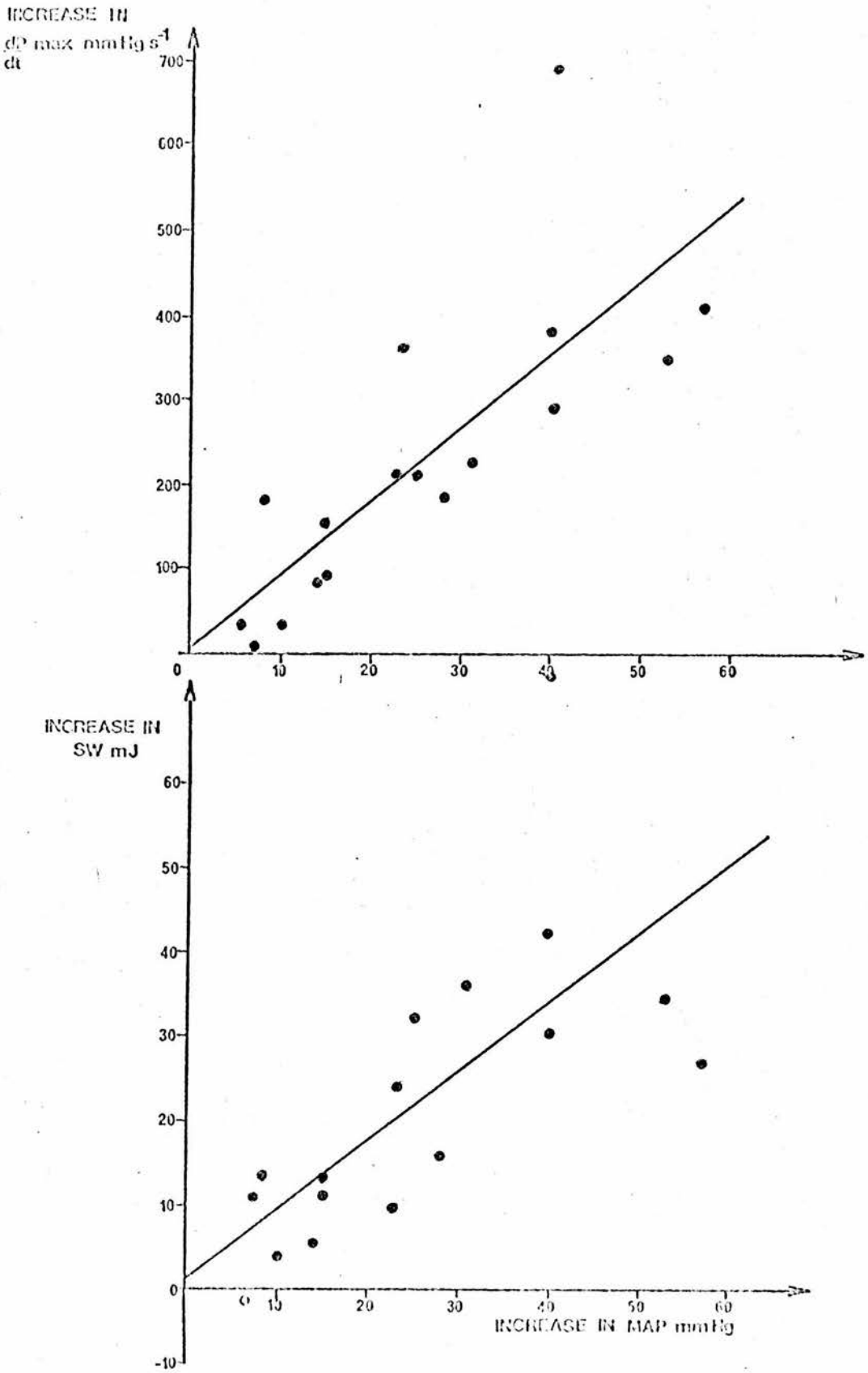


Figure 5. The effect of MAP on $\frac{dP}{dt}$ max and SW. A. The relationship betwee increase in MAP and the change in $\frac{dP}{dt}$ max. The mean lowest $\frac{dP}{dt}$ m was 1571 mmHgs^{-1} (SD± 24). The regression equation is Y=8.6±1.8 B The relationship between the increase in MAP and the change in The mean lowest SW was 65.1 mJ (SD±16). The regression equation is Y = 0.8x+1.8.

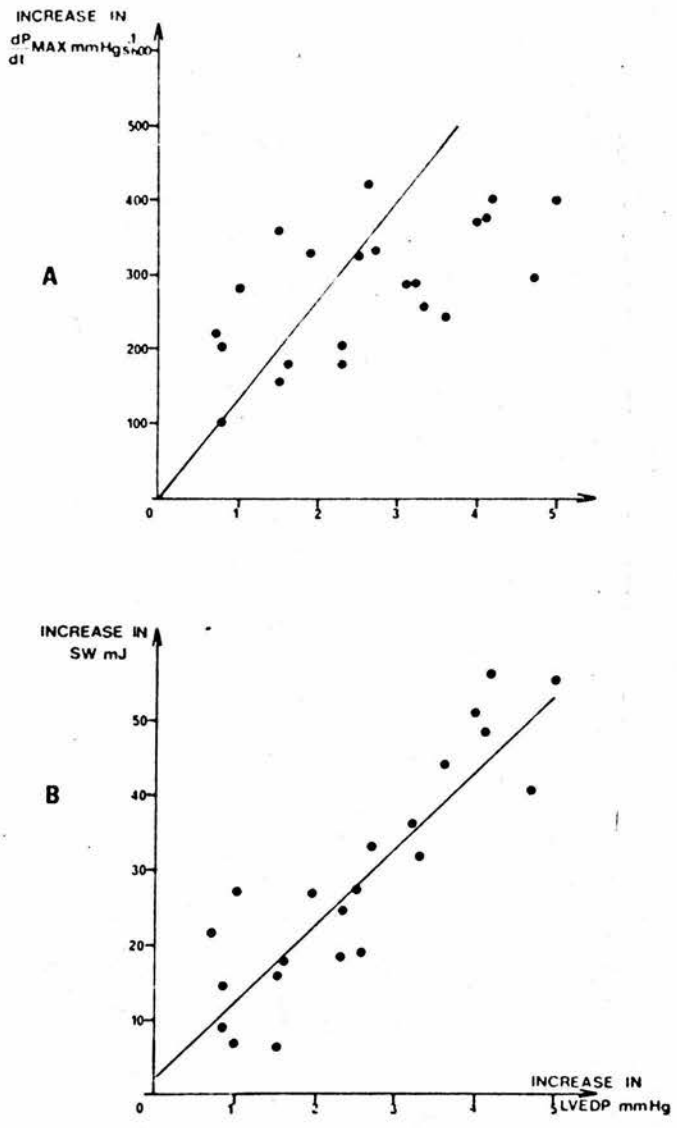


Figure 6. The effect of LVEDP on $\frac{dP}{dt}$ max and SW.

A. The relationship between the increase in LVEDP and the change in $\frac{dP}{dt}$ max. The mean lowest value for $\frac{dP}{dt}$ max was 1165 $mmHgs^{-1}$ (S.D. $\pm$ 243). The regression equation is $Y = 134x$.

B. The relationship between the increase in LVEDP and the change in SW. The mean lowest value for SW was 22.9 mJ (S.D. $\pm$ 10.9) The regression equation is $Y = 10.1x + 2.6$,

In each animal an increase in LVEDP produced an increase in $\frac{dP}{dt}$ max. This is not a linear relationship, the increases in $\frac{dP}{dt}$ max being greater in the lower range of LVEDP. The best straight line which passes through the observed points and the origin was calculated to provide an estimate of the effect of a rise in LVEDP on $\frac{dP}{dt}$ max. The slope of this line indicates a rise in $\frac{dP}{dt}$ max of 134 mm Hg s^{-1} for a rise in LVEDP of 1 mm Hg. This is probably an underestimate at low LVEDP.

The relationship between LVEDP and SW, however, does seem to be linear and the slope of the calculated regression line indicates an increase in SW of 10 mJ for an increase in LVEDP of 1 mm Hg.

The relationship between change in $\frac{dP}{dt}$ max and SW during infusion of noradrenaline.

An example of the records obtained during a single infusion of noradrenaline is shown in Figure 7 . During this particular infusion HR was held constant at 139 beats/min, MAP at 68 mm Hg, and LVEDP at 3.2 mm Hg. $\frac{dP}{dt}$ max rose from 1631 to 2617 mm Hg s^{-1}, SW rose from 54.7 to 85.6 mJ, and SV from 3.9 to 5.4 mls.

The results of 32 similar tests in sixteen dogs are shown in Figures 8&9 and the data from which these graphs were drawn is tabulated in table D . The mean HR was 182 beats/min (SD^{+} 22), the mean MAP was 85 mm Hg (SD^{+} 10) and the mean LVEDP was 2.5 mm Hg.

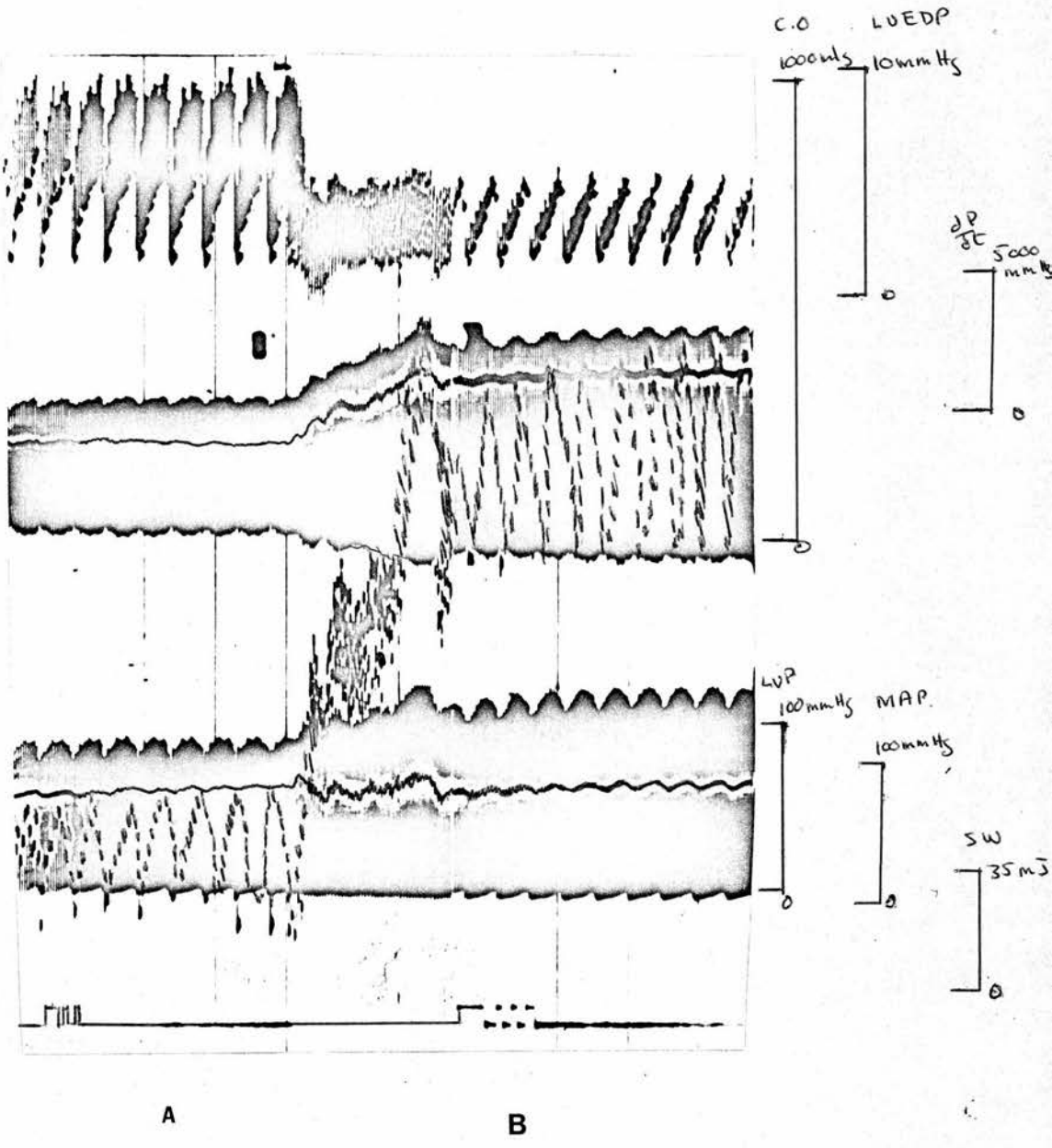


Figure 7. Example of records obtained after a single infusion of noradrenaline. Panel A, control. Panel B, noradrenaline infused at 0.19 ug/kg/min. From above downwards the traces are: LVEDP, $\frac{dP}{dt}$, C.O., LVP, MAP, SW.

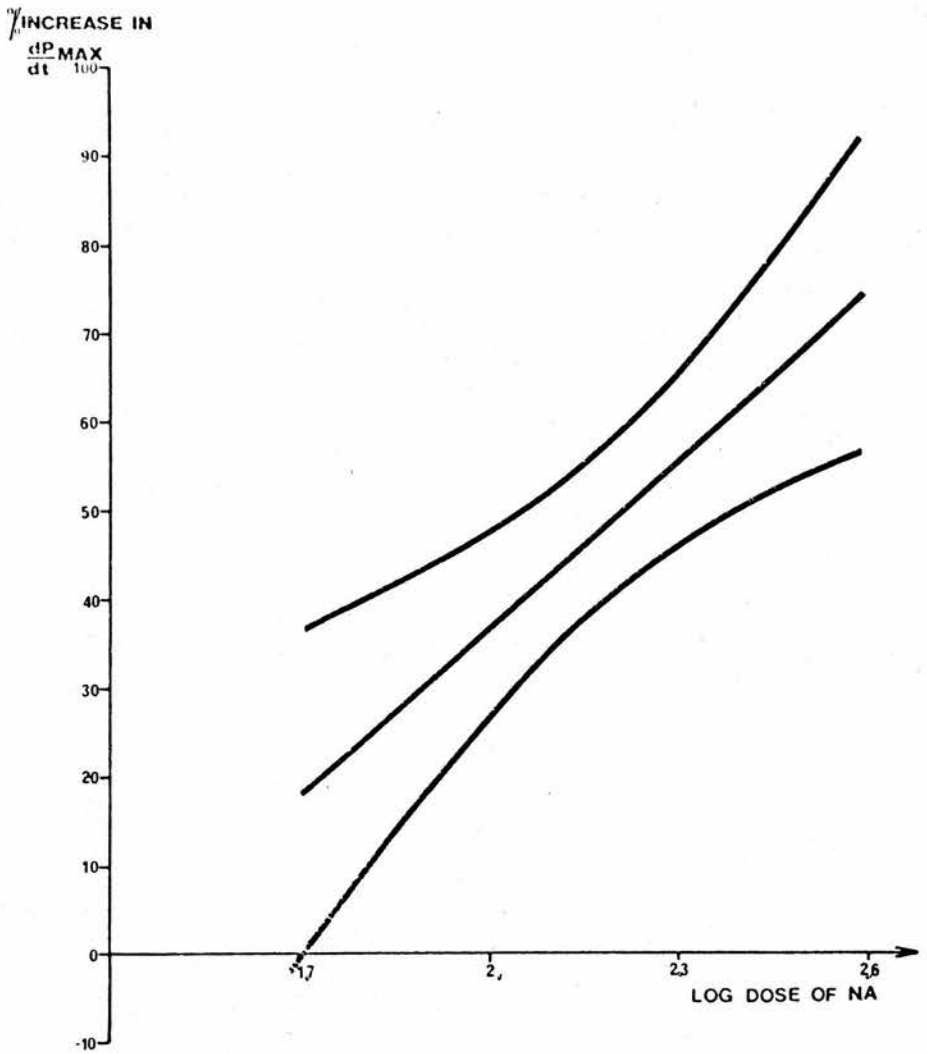


Figure 8. The effect of noradrenaline infusion on $\frac{dP}{dt}$ max.

Dose response curve showing the effect of log. rate of infusion of noradrenaline on the % change in $\frac{dP}{dt}$ max

The mean resting value for $\frac{dP}{dt}$ max was 1917 mm Hgs^{-1} (S.D.$\pm$ 383). The regression equation is $Y=62.8_X - 89$.

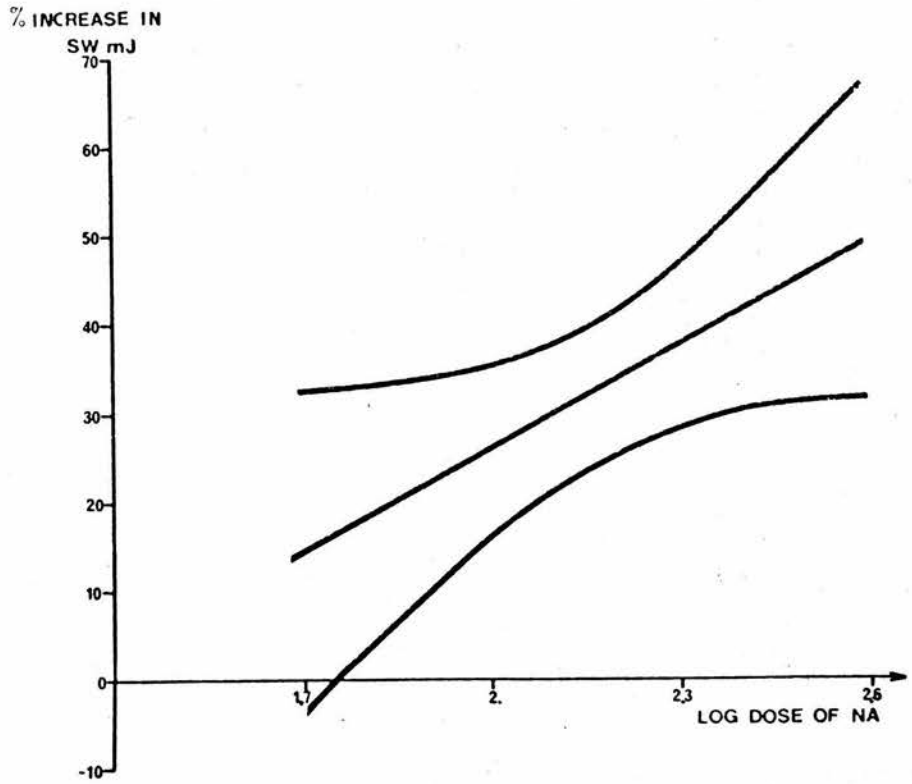


Figure 9. The effect of noradrenaline infusion on SW.

Dose response curve showing the effect of log. rate of infusion of noradrenaline on the % change in SW. The mean resting value of SW was 56.6 mJ (S.D. $\pm$ 19.2). The regression equation is Y = 38.2x - 50.6.

During each test HR was held constant, MAP and LVEDP were held constant within $\pm$ 4 mm Hg and $\pm$ 0.4 mm Hg respectively.

The regression lines with 95% confidence limits are shown. Increasing the rate of noradrenaline infusion produced dose dependant increases in $\frac{dP}{dt}$ max ($P<0.005$) and in SW ($P<.05$). In Figure 10 the rate of noradrenaline infusion is related to the % change in approximate SW, calculated from the formula SW = SV(MAP-LVEDP), and the 95% confidence limits have been drawn. Noradrenaline increased approximate SW. Although the increases in approximate SW were significant, the slope of the increase with increasing dose of noradrenaline was not significant at the 5% level.

In seven out of the thirty two tests, an increase in $\frac{dP}{dt}$ max was recorded without an increase in either computed or approximate SW. In all of these cases, although noradrenaline increased the rate of development of pressure, peak systolic pressure was not increased (table D).

DISCUSSION

The effect of an increase in heart rate.

From the results shown in Figures 3&4 it can be seen that as the HR rises, $\frac{dP}{dt}$ max continues to increase, although SW and cardiac power fall above an optimum HR. These effects were associated with either no change or a slight fall in peak systolic pressure and a large decrease in the duration of contraction and

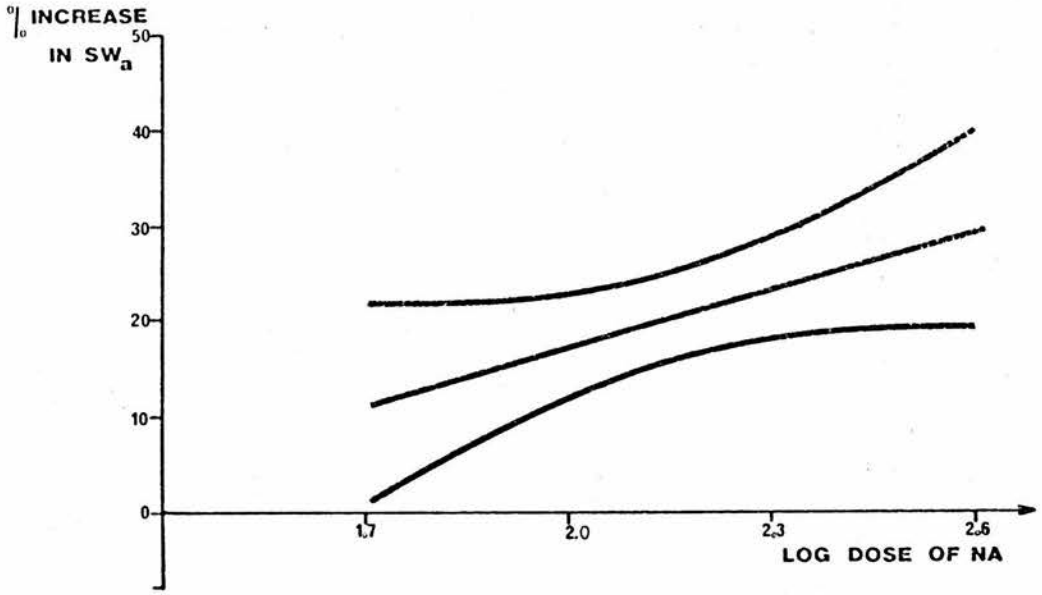


Figure 10. The effect of noradrenaline infusion on approximate SW (SWa), i.e. SV (MAP-LVEDP). Dose response curve Showing the effect of log. rate of infusion of noradrenaline on the % change in SWa. The mean resting value of SWa was 40.2 mJ (S.D. $\pm$ 12). The regression equation is Y =20.2x - 23.

systole (Figure 11Table E). Thus at high heart rates, a further increment in HR increases $\frac{dP}{dt}$ max whereas peak systolic pressure is unchanged or decreased slightly. This effect of an increase in HR on myocardial force of contraction has previously been reported in isolated mammalian atria (Kruta and Stejskalova 1960) and in papillary muscle (Sonnenblick 1962: Blinks and Koch-Weser 1961). In the heart in situ, when changes in HR were investigated, ventricular function curves have shown no change (Mitchell et al 1963) or a decrease in the strength of contraction (Berglund et al 1958).

It has been postulated that the decrease in tension occurring in isolated cardiac muscle, at higher frequencies is secondary to a shortening of the duration of the contraction (Koch-Weser& Blinks 1963: Sonnenblick 1965). Over a wide range of rate increments the effect of the increasing degree of activation of the contractile elements on the amount of tension or pressure developed more than offsets the affect of the decreasing duration of contraction. At high heart rates, however, changes in duration become more important and may determine the direction of a change in the strength of contraction.

This effect would explain why at high rates we found that $\frac{dP}{dt}$ max still increased, although peak systolic pressure, SW and cardiac power decreased above an optimum HR. Similar observations by Siegel et al (1964) and Covell et al (1966) led these authors to conclude that the ventricular function curve was unable to assess alterations in the contractile state of the heart which

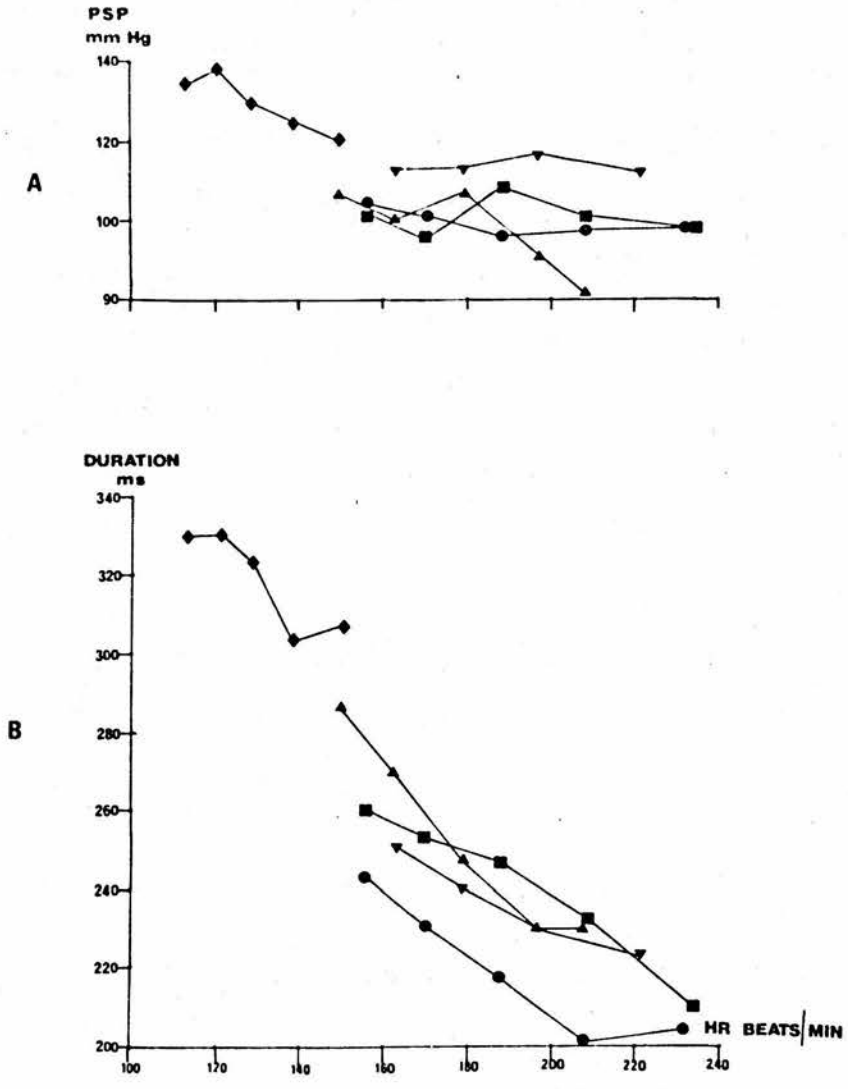


Figure 11. The effect of increasing heart rate on A, peak systolic pressure (PSP): and B, the duration of contraction Each symbol represents the results obtained in one dog.

were reflected primarily by a change in the velocity of contraction. Siegel et al (1964) further concluded that changes in contractility were characterised by a change in the velocity dependant aspects of contraction and therefore the best index of this aspect of contraction was most reliable as an index of contractility.

However, as previously discussed in the introduction, a complete description of the contractile state of the heart requires the use of four variables, namely, the tension in the contractile component, its velocity of shortening, the muscle length and the time after excitation. Changes in the velocity dependant aspects of contraction reflect only one facet of the contractile state of the heart and indices of this aspect of contraction, e.g. $\frac{dP}{dt}$ max, can only validly be used to reflect a change in the force-velocity relationship, i.e. a change in contractility when force of contraction and velocity of contraction are changed in parallel.

As seen in Figure 3 and 11, the effect of increasing HR is an example of a situation where force and velocity are changed in different directions. In such cases, if only one index, namely $\frac{dP}{dt}$ max was used to assess a change in the force-velocity-length relations an incomplete description of the inotropic intervention would be obtained.

The effect of an increase in afterload.

Our results confirm that an increase in MAP with the heart

rate and LVEDP held constant is accompanied by an increase in SW and $\frac{dP}{dt}$ max. It is a fundamental property of muscle that the work done in a contraction is partly determined by the afterload. If afterload is increased, the work done increases from zero, when the external resistance to shortening is zero, as a function of afterload up to a maximum value and then decreases to zero, when the afterload is too great to allow shortening. This phenomenom does not by definition include an increase in contractility, but it has been suggested that, in the whole heart the response to an increase in aortic pressure also includes a positive inotropic effect (Von Anrep 1912, Sarnoff et al 1960b).

Our results support the hypothesis that an increase in MAP is accompanied by a small increase in contractility, as indicated by an increase in the value of $\frac{dP}{dt}$ max of the left ventricle. $\frac{dP}{dt}$ max, if it occurs within the isovolumic phase of contraction is not dependant on afterload (Siegel et al 1964). There has been some dispute in the literature about the exact point during the isovolumic contraction when $\frac{dP}{dt}$ is maximal. It has been suggested that if $\frac{dP}{dt}$ max does not occur before the aortic valve opening, then a change in aortic diastolic pressure may change $\frac{dP}{dt}$ max, because the opening of the valve would then normally limit the magnitude of $\frac{dP}{dt}$ max.

Examination of our records, however, showed that in all but three tests, $\frac{dP}{dt}$ max occurred before the time of aortic

valve opening, as indicated by the initiation of aortic flow. In the tests in which $\frac{dP}{dt}$ max did not occur before valve opening, the aortic pressures were the lowest recorded in each of the series of tests. This suggests that the increase in $\frac{dP}{dt}$ max which occurred when afterload was raised from a very low level was partly secondary to the fact that at the lower afterload, shortening had occurred prior to the achievement of $\frac{dP}{dt}$ max, and therefore the value of $\frac{dP}{dt}$ max was lower than it would have been otherwise.

The mechanism underlying this positive inotropic effect is unknown. Starling (1918) attributed this effect mainly to the improved myocardial perfusion, following the increased coronary perfusion pressure. The studies of Rosenblueth (1959b), however, have shown an essentially similar response in the right ventricle, while the coronary arteries were independently perfused. Local release of catecholamines is unlikely to be involved since the response has been demonstrated after propranalol (Clancy et al 1968) and in the reserpinised animal (Blinks and Koch-Weser 1963). Rosenblueth et al (1959) and Sarnoff et al (1963) suggested that myocardial contractility is somehow determined by the amount of work performed by the heart in preceding beats. Sarnoff showed a net loss of K^+ during an increase in aortic pressure and related this to an altered metabolic state which subsequently influenced the following beats.

A conclusion about the mechanism involved in the increased

contractility following a rise in MAP, cannot be drawn from our results. Quantitative assessment of this effect shows that the changes in $\frac{dP}{dt}$ max and in SW are proportional to the changes in MAP. For a rise in MAP of 10 mm Hg, $\frac{dP}{dt}$ max rose by 86 mm Hg s^{-1} and SW rose by 8 mJ. Although these changes are small compared to the inotropic changes associated with noradrenaline, they are large enough to necessitate the control of MAP, if one uses SW or $\frac{dP}{dt}$ max to measure inotropic changes.

The effect of an increase in preload.

The importance of the influence of initial fibre length or end diastolic volume on ventricular performance has been appreciated for many years. Although it has been generally accepted that stroke work increases as LVEDP is increased, there has been some disagreement recently as to whether or not $\frac{dP}{dt}$ max is influenced by changes in LVEDP. Our results confirm the findings of Wallace et al (1963). Increasing LVEDP, while HR and MAP are held constant produced increases in $\frac{dP}{dt}$ max and in SW (Figure 6). In isolated muscle, the rate of force development has been shown to be linearly related to initial muscle length. Since the rate of force development in the ventricle wall is related to $\frac{dP}{dt}$ by the Laplace relationship i.e.

$$\frac{dP}{dt} = \frac{\frac{dF}{dt}}{R}$$

where R is the radius of the ventricle, an increase in end

diastolic volume, over the range which we studied, must have had a greater effect on $\frac{dF}{dt}$ than on the radius of the ventricle. Consequently $\frac{dP}{dt}$ max was increased.

For a rise in LVEDP of 1 mm Hg, $\frac{dP}{dt}$ max rose by 134 mm Hg s^{-1} and SW by 10 mJ. These results are in disagreement with the findings of Furnival et al (1970). They found that $\frac{dP}{dt}$ max was independent of preload, and on this basis concluded that $\frac{dP}{dt}$ max could be used to indicate an inotropic effect even in the presence of changes in the end-diastolic volume of the ventricle. The discrepancy between these results may partly be explained by the difference in the range of end diastolic pressures studied. Furnival et al observed the effect of a mean increase in LVEDP of 5 mm Hg from the mean control level of 5 mm Hg. Whereas our mean increase was 5 mm Hg from a control level of 1 mm Hg. We observed that the increments in $\frac{dP}{dt}$ max became smaller at higher end diastolic pressures and as Furnival et al worked over a higher range of pressures this may explain why they did not observe consistant increases in $\frac{dP}{dt}$ max.

Our results support the conclusions drawn by most workers that LVEDP must be maintained constant, when $\frac{dP}{dt}$ max or SW are used to assess inotropic changes in the heart.

The inotropic effect of catecholamines

In the heart in situ, Wiggers (1927) showed that the effect

of catecholamines on the contractile state of the heart is characterised by two distinct alterations in the contractile process. Catecholamines increase the rate of rise of pressure in the ventricle and decrease the duration of systole. These changes indicate that the contractile component must be shortening faster and that the duration of activity must be reduced. The increase in $\frac{dP}{dt}$ max reflects that catecholamines alter the force-velocity relationship of cardiac muscle. Noradrenaline has been shown to increase Vmax, the maximum velocity of shortening, in cat papillary muscle (Sonnenblick 1962) but its action on Po, the maximum force generating capacity of muscle, has not yet been determined. Brady (1967) observed that catecholamines did not alter the contracture tension of frog muscle in high potassium chloride solution. The failure of catecholamines to alter the contracture tension suggests that catecholamines do not affect a mechanism which necessarily governs peak tension development. He concluded that the consistant effect of catecholamines was to increase the rate of tension development rather than necessarily its maximum amplitude. This conclusion was supported by his finding in rabbit papillary muscle, contracting isometrically, that if the resting muscle length and the stimulation frequency are chosen for maximal contraction, then addition of a catecholamine will only increase the rate of rise and fall of tension and not the peak tension. Without the presence of another inotropic intervention, however, catecholamines usually increase the peak tension. An increase in $\frac{dP}{dt}$ max would produce

an increase in the peak systolic pressure developed in the ventricle, unless this effect is offset by a big decrease in the duration of contraction.

The results of the present study indicate that as the rate of infusion of noradrenaline is increased, under conditions in which HR, MAP and LVEDP are held constant at their preinfusion levels, there are proportional increases in both $\frac{dP}{dt}$ max and SW. Furnival et al (1970) working under similarly controlled conditions did not obtain consistant changes in SW. Three factors may explain the discrepancy between their results and ours.

1. They calculated SW by the usual approximation. The true external SW is the integral with respect to time of the instantaneous power i.e. the product of pressure and flow and not the product of the integrals with time of the out of phase peaks of pressure and flow. This approximation tends to underestimate the changes induced by catecholamines, since although MAP has been held constant, systolic pressure is raised. If our values of SW are compared with those obtained, from the same records, using the approximation it is found that on infusion of noradrenaline the changes in computed SW are usually greater than the changes in approximate SW (table D). The difference between the two values of the changes in SW is found to be directly related to the change in peak systolic pressure, as is shown in Figure 12 .

2. Furnival et al maintained MAP by clamping the descending aorta

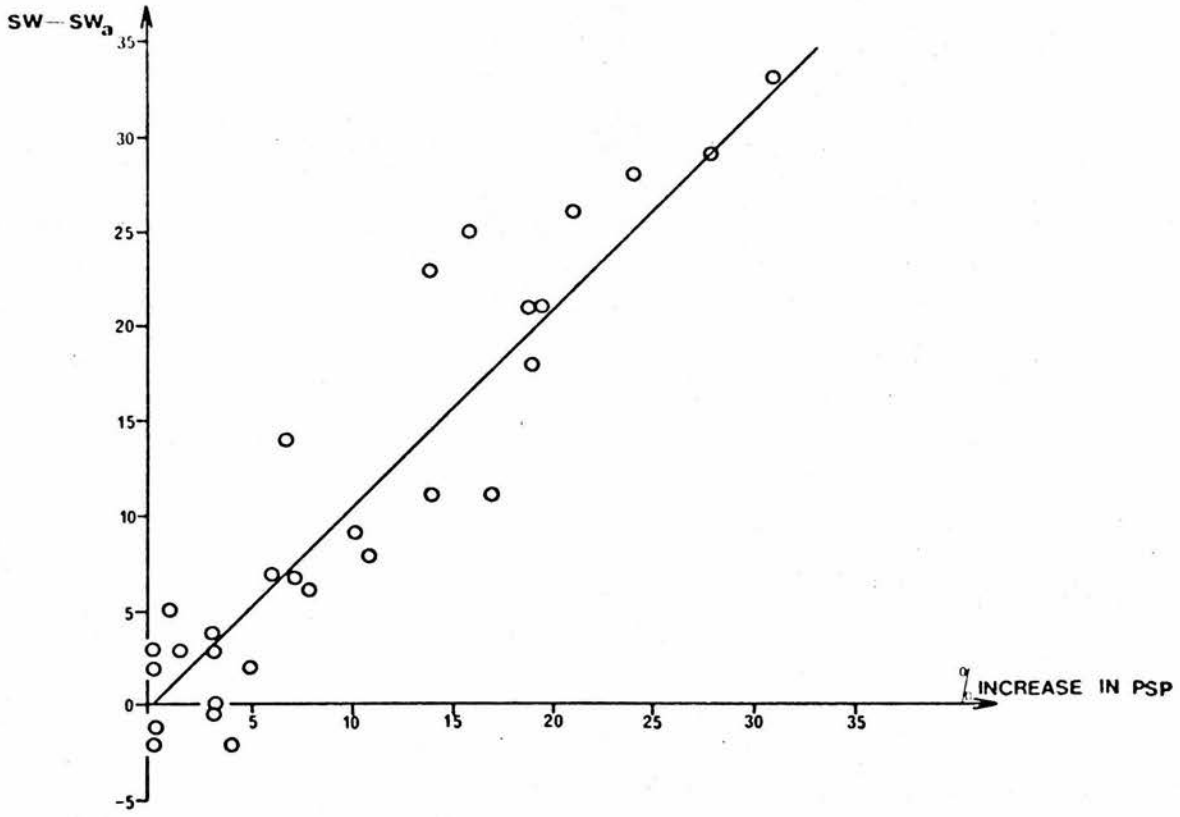


Figure 12. The relationship between the change in peak systolic pressure and the discrepancy between the computed change in SW and the conventionally calculated change in SW (SWa). The mean resting value of systolic pressure was 118 mm Hg (S.D. $\pm$ 16).

during isoprenaline infusions. This intervention by reducing the compliance of the arterial tree as well as increasing its resistance may reduce the phase angle between peak pressure and peak flow, increasing the SW by a component missed by the approximation.

3. In the experiments of Furnival et al, for the measurement of $\frac{dP}{dt}$ max and SW, the hearts were paced at a rate just below that producing pulsus alternans. The existance of an upper limit to the contractile response has been noted by a number of workers (Blinks and Koch-Weser 1963). Under the experimental conditions of such a high heart rate, the heart is already working at the upper limit of its contractile strength and the influence of nonspecific limits on the contractile response may have interfered with the action of catecholamines.

For this reason, in order to obtain a useful concentration response curve of an inotropic drug, the heart should be able to contract under conditions which allow it to respond with changes in the strength of contraction that are proportional to the underlying changes in the degree of activation. This is not the case, if the control strength of contraction is already near its maximum value, as at a high heart rate or in high extra cellular calcium concentration.

That the positive inotropic effect of catecholamines may be greater at intermediate than at high heart rates has previously

been shown in isolated muscle preparations. Kruta and Zadina (1938) showed in isolated guinea pig atria, that as the frequency of contraction was increased above an optimum frequency, the effect of adrenaline on the amplitude of contraction was decreased. In kitten atrial strips, it has also been shown that catecholamines flatten the interval-strength relationship at a high level causing the strength of contraction to be nearly uniform over a range of intermediate and high frequencies. (Koch-Weser et al 1963) . Catecholamines have also been shown to have no positive inotropic effect, as measured by a change in developed tension, on the isometric contracture tension of cat papillary muscle in a calcium rich solution (Kavaler and Morad 1966). In some experiments, a small decrease in tension was observed after the addition of adrenaline this effect being attributed to the decrease in the duration of the contraction produced by the drug.

From the results shown in Figure 11 , it can be seen that in the intact heart, which is being stimulated at a rate just below that producing below pulsus alternans, the duration of contraction is much reduced and the greatest tension which the muscle is capable of generating in that time, is probably being developed. The infusion of a catecholamine under these circumstances may be unable to increase the peak systolic pressure or to increase the external work done although $\frac{dP}{dt}$ max is still increased. In some experiments we repeated the infusion of noradrenaline at a rate just below that producing pulsus alternans and in some cases,

as in Figure 13 , we were able to illustrate such an effect. This is another example of a situation in which the force and velocity of contraction are not changed in parallel. These effects may explain why in some tests with noradrenaline, at high heart rates, we observed only an increase in $\frac{dP}{dt}$ max without an increase in SW, and also why Furnival et al found changes in SW to be an unreliable guide to the inotropic effect of catecholamines.

We have confirmed that $\frac{dP}{dt}$ max is more consistantly affected by catecholamines than is external work. We have found, however, that in hearts which are not driven near their limits of performance, SW is affected in a consistant and dose-dependent manner by catecholamines. Stroke work calculated without approximations, under conditions of controlled heart rate, aortic pressure and filling pressure is a reliable index of inotropic changes and has the merit of directly assessing the performance of the heart as a pump. Furthermore under conditions in which $\frac{dP}{dt}$ max and stroke work consistantly diverge it becomes misleading to regard changes in $\frac{dP}{dt}$ max as inotropic changes.

We have shown that such a situation arises when divergent changes in the velocity and force of contraction occur, e.g. at high heart rates and also when the inotropic effects of catecholamines are observed in the heart which is already working near the upper limit of its contractile strength. If one attempts to relate the action of inotropic interventions to the fundamental properties of muscle, i.e. according to their effects on the time

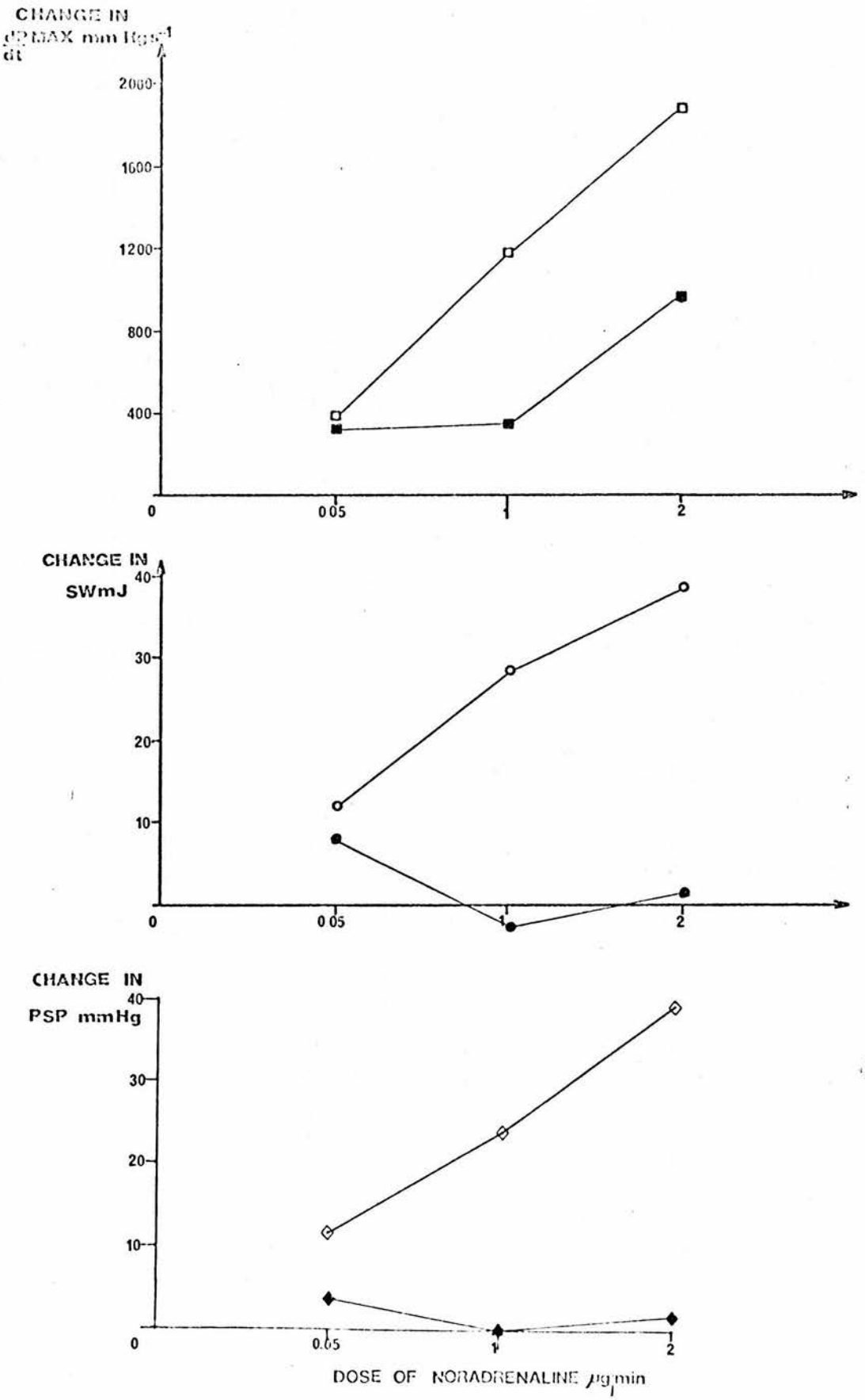


Figure 13. Dose-response curves for noradrenaline at two heart rates in one preparation.
Squares represent changes in $\frac{dP}{dt}$ max Circles represents changes in SW. Diamonds represent changes in PSP. Open symbols represen changes at HR 179 beats/min. Closed symbols represent changes a HR 208 beats/min.

course of contraction, as well as their effect on peak tension and rate of development of tension, it would perhaps be more appropriate to revive the term 'klinotropic effect', as suggested by Reiter (1972) to denote a change in the rate of development of tension.

S E C T I O N 2 .

The haemodynamic effects of propranalol and sotalol.

Table 1 shows the basal control levels of the measured cardiac variables. Heart rate was the only parameter, in which there was a difference between the control levels in the dogs given sotalol and those given propranalol. Preliminary investigations had shown that sotalol but not propranalol always caused a large fall in heart rate. In order that we could observe the effects of sotalol at constant heart rate, in subsequent experiments using sotalol, we paced the hearts at the lowest rate which they could follow.

Effects of sotalol and propranalol on normal dogs.

After each dose of the drug, HR was held constant at its control value, MAP and LVEDP were held to within $\pm$ 5 mm Hg and $\pm$ 0.5 mm Hg respectively, except in 2 tests when MAP fell 10 mm Hg.

Administration of sotalol, 1-16 mg/kg, caused a decrease in $\frac{dP}{dt}$ max and in SW (Figure 14 : table F). The initial dose of sotalol produced a mean reduction in $\frac{dP}{dt}$ max and in SW of 9%. Subsequent doses produced only minor further changes. A dose of 32 mg/kg usually depressed the heart such that it was no longer

TABLE 1 .

Control levels of the measured cardiac variables.

Variable	NORMAL DOGS		RESERPINIZED DOGS		PRACTALOL-TREATED DOGS
	Sotalol n = 4	Propranalol n = 3	Sotalol n = 3	Propranalol n = 4	Sotalol n = 3
Heart Rate (beats/min)	143 ± 25	179 ± 7	130 ± 7	159 ± 16	146 ± 5
Mean Aortic Pressure (mm Hg)	75 ± 3.5	77 ± 4	80 ± 11	79 ± 5	94 ± 5
LVEDP (mm Hg)	3.6 ± 1.1	2.6 ± 0.4	2.4 ± 0.6	3.7 ± 0.9	1.0 ± 0.3
$\frac{dP}{dt}$ max ($mm\ Hg\ s^{-1}$)	1444 ± 392	1875 ± 506	1640 ± 508	1490 ± 130	2219 ± 186
SW (mJ)	48.1 ± 12.5	49.6 ± 9.9	89.4 ± 24.5	55.4 ± 7.3	87.7 ± 2.1

Figures quoted are the means of absolute values (± SD) recorded prior to administration of either sotalol or propranalol in the indicated (n) number of normal, reserpinized or practalol treated dogs.

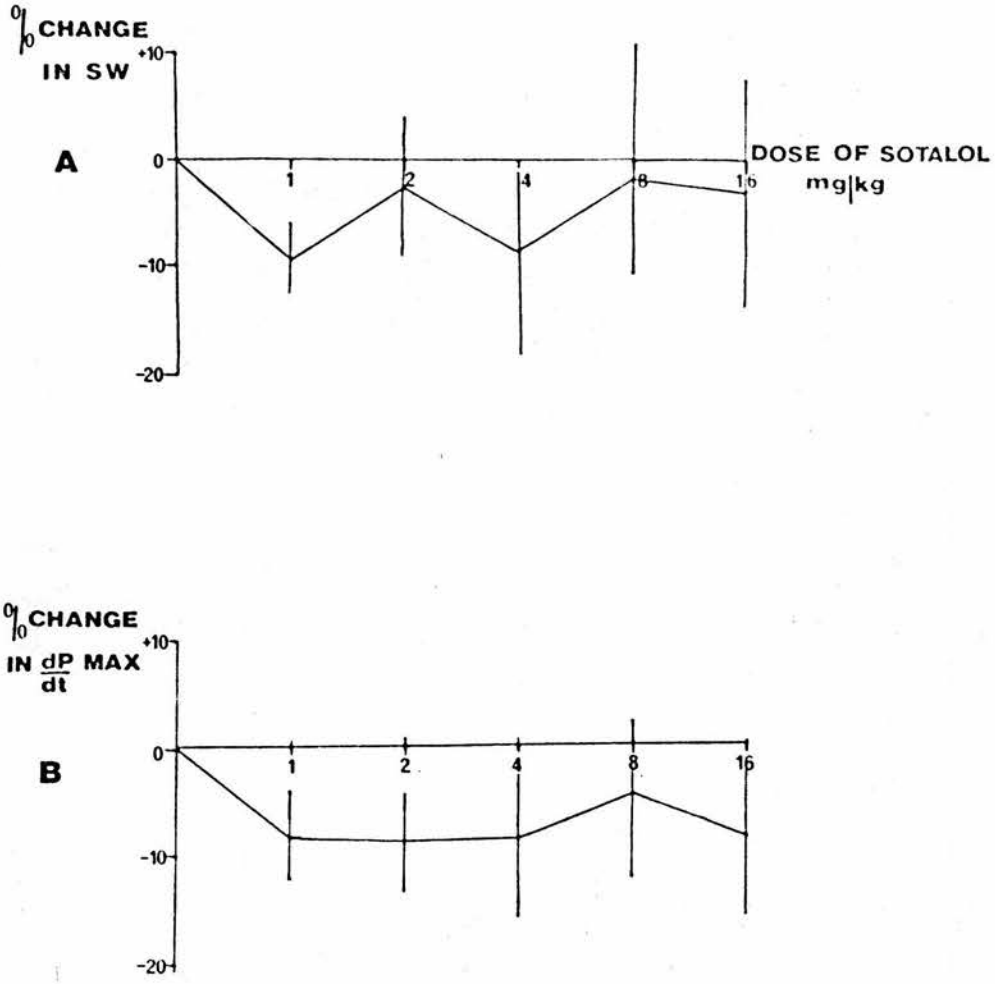


Figure 14. The effect of cumulative doses of sotalol on $\frac{dP}{dt}$ max and SW in 4 dogs, while HR, MAP and LVEDP were held constant

A. The rel ationship between the total dose of sotalol and the % change in SW from control $\pm$ 1SEM. The mean resting value of SW was 48.1 mJ (S.D.$\pm$ 12.5)

B. The relationship between the total dose of sotalol and the % change in $\frac{dP}{dt}$ max from control $\pm$ 1SEM. The mean resting value of $\frac{dP}{dt}$ max was 1444 $\pm$ 392.

possible to maintain MAP and LVEDP at their control valves. There was no difference in the response of SW and $\frac{dP}{dt}$ max to sotalol.

The effects of sotalol on the duration of the ventricular pressure pulse, the duration of systole and the duration of the Q-T interval of the ECG are shown in Figure 15 and table F . Dose dependant increases in these three parameters were observed. At each dose level, the biggest increase in duration was observed in the ECG.

Propranalol, 0.1 - 3.2 mg/kg, decreased $\frac{dP}{dt}$ max and SW (Figure 16 table G). The reductions in both parameters following propranalol were greater than those observed after sotalol, the initial mean reduction being of the order of 20%. Further increases in the dose of propranalol produced no further reduction until a cumulative dose of 0.8 mg/kg. Doses greater than 3.2 mg/kg produced a further big depression of the myocardium. Most animals did not survive a dose of 6.4 mg/kg.

Table G shows the effect of propranalol on the duration parameters. Propranalol, in doses up to 3.2 mg/kg did not produce any increase in the duration of the Q-T interval of the ECG. In one out of three experiments the initial dose of propranalol produced an increase in the duration of the ventricular pressure pulse. In this particular experiment the initial dose of propranalol had caused a very large depression of the heart $\frac{dP}{dt}$ max decreased by 40% of its control valve. A similar increase in

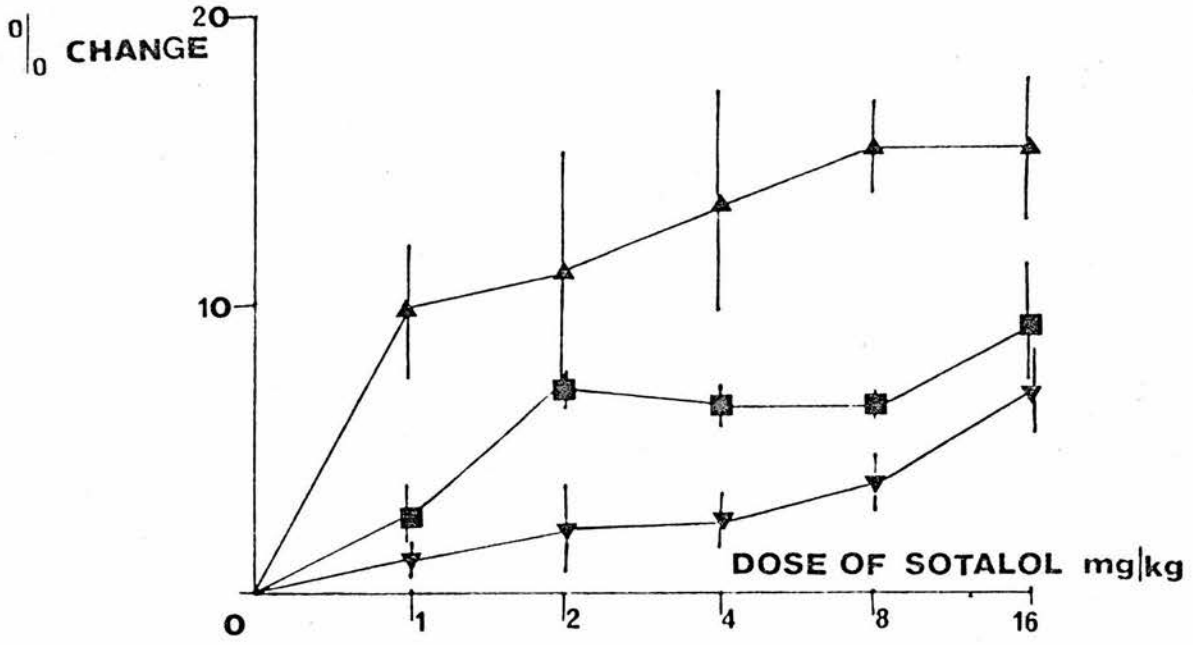


Figure 15. The effect of cumulative doses of sotalol on the % change in the duration of the ECG ▲, the duration of the ventricular pressure pulse ■, and the duration of systole ▼ The mean control value of the duration of the ECG was 223 ms (SD ± 26), of the ventricular pressure pulse was 282 ms (SD± 30), and of systole was 235 ms (SD ± 18).

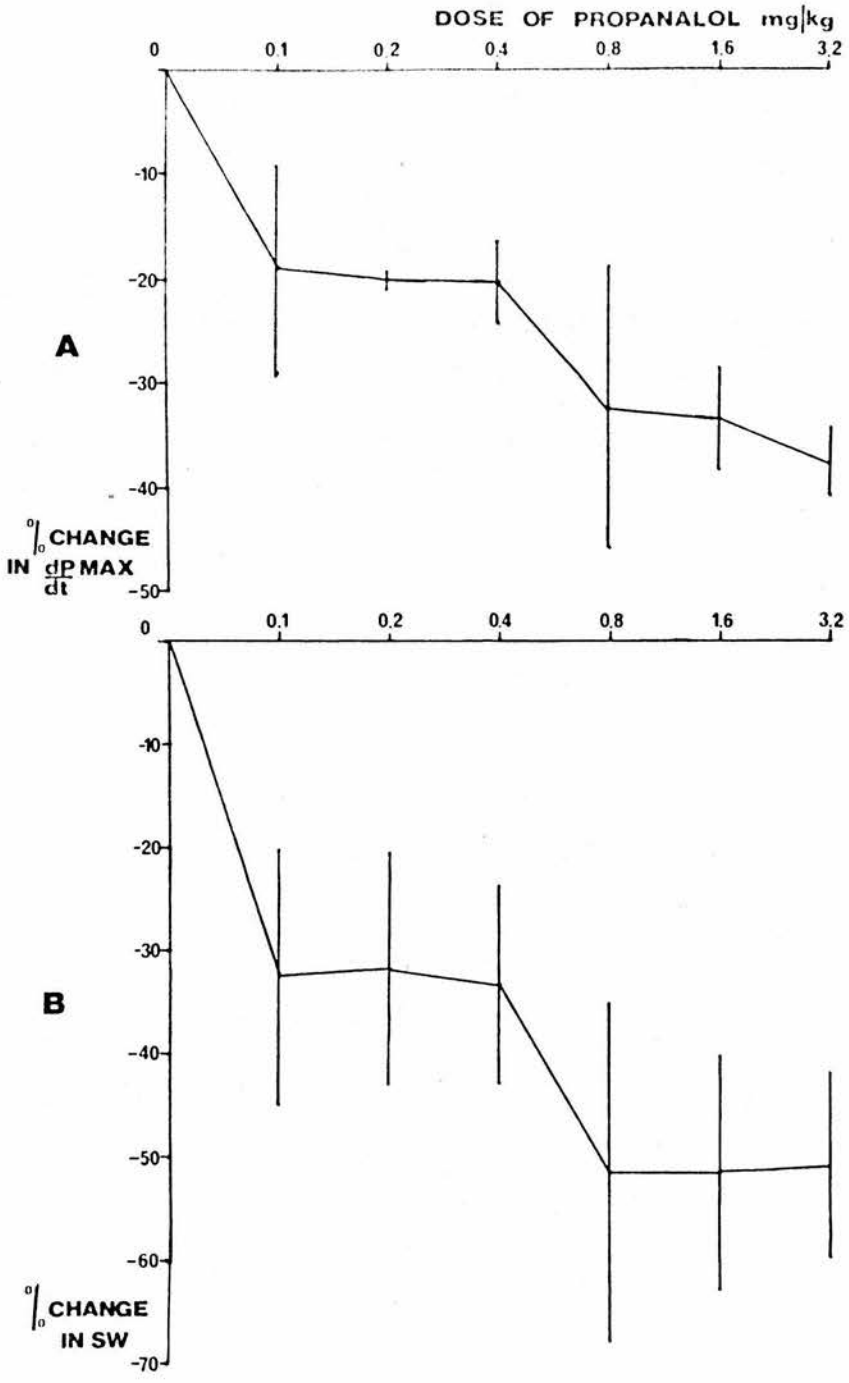


Figure 16. The effect of propranalol on $\frac{dP}{dt}$ max and SW in 3 normal dogs. in which HR, MAP and LVEDP were held constant.

A. The relationship between the total dose of propranalol and the % change from control in $\frac{dP}{dt}$ max $\pm$ 1SEM.

B. The relationship between the total dose of propranlol and the % change from control in SW $\pm$ 1SEM.

the duration of contraction was observed, without a corresponding change in the duration of the ECG, when a dose of 6.4 mg/kg also produced a large myocardial depression.

Effects of sotalol and propranabl on reserpine pretreated dogs.

All these animals appeared lethargic and depressed 24 hours after the administration of reserpine . The initial dose of anaesthetic required was usually reduced to a half of that required for normal animals. The response to tyramine was measured at the beginning of each experiment to test the degree of depletion of catecholamines in the heart. MAP was increased by a mean of 6.8% and HR by a mean of 1.7%. These responses demonstrate that a substantial depletion of catecholamines had been produced by the reserpine. We found the reserpinized preparations to be very labile and prone to myocardial depression. Subsequently it was difficult to hold MAP and LVEDP at their control values. After the administration of each dose of the drug HR was held constant and LVEDP held to within $\pm$ 0.4 mm Hg of the control value MAP was maintainted to within $\pm$ 5 mm Hg in most tests, although in some cases a change of 5 - 10 mm Hg was unavoidable.

Figure 17 and table H shows the pooled data from three reserpinized dogs after administration of sotalol, 1 - 32 mg/kg. In one experiment, sotalol caused a 40 - 50% depression of contractility over the whole dose range. This effect was

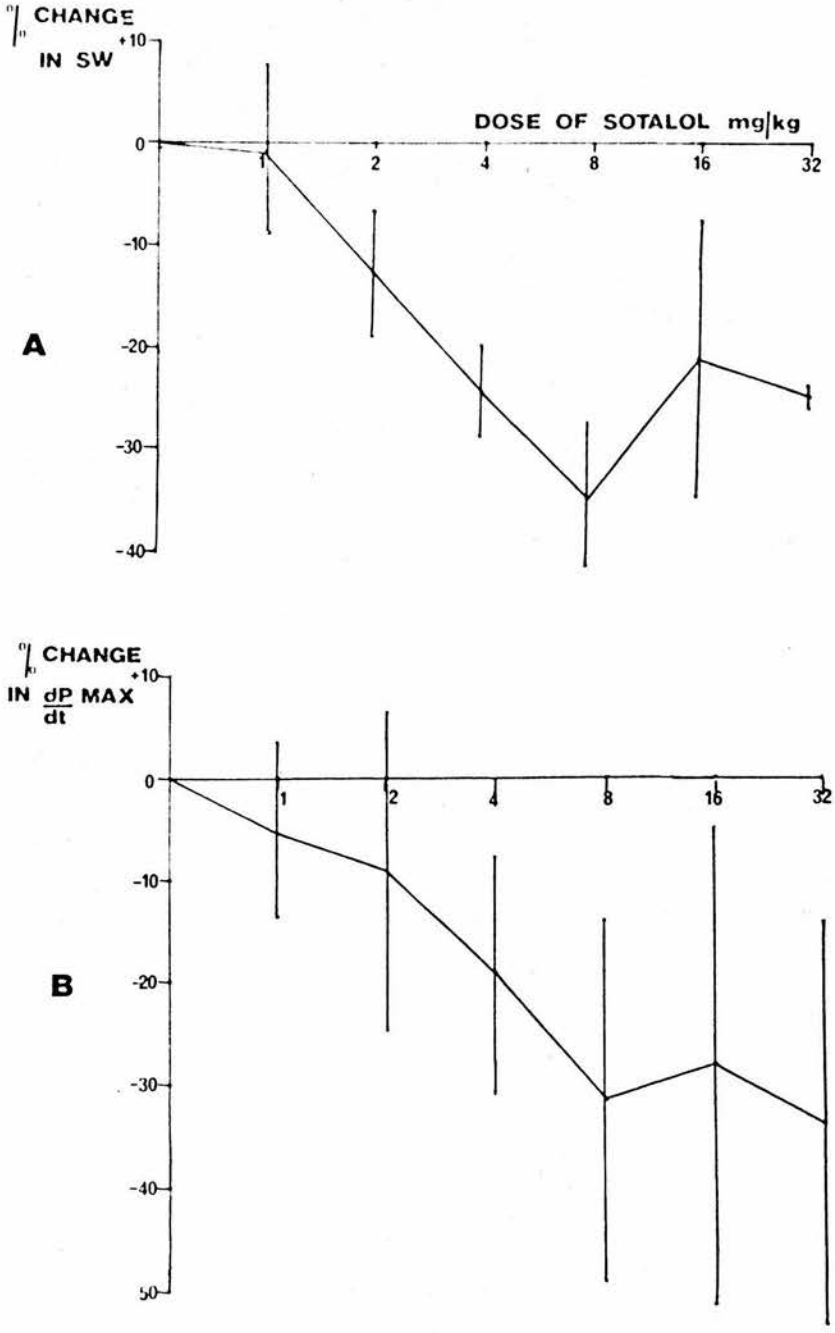


Figure 17. The effect of sotalol on 3 reserpinized dogs. While HR, MAP and LVEDP were held constant.

A. The relationship between the total dose of sotalol and the % change in SW from control ± 1SEM.

B. The relationship between the total dose of sotalol and the % change from control in $\frac{dP}{dt}$ max ± 1SEM.

inconsistant with the results from the other two experiments. Inclusion of this experiment into the pooled data, however, would make our results indicate that sotalol has apparently a greater negative inotropic effect in reserpinized than in normal dogs.

The effects of sotalol on the duration parameters are shown in Table H . In the inconsistant experiment sotalol did not cause any change in the duration of the Q-T interval of the ECG. The increase in the duration of the ventricular pressure pulse observed in this experiment was associated with a large fall in the contractile state of the heart. In the other reserpinized dog, which received the full dose range, we observed a consistant increase in the duration of ECG and of the ventricular pressure pulse.

The effect of increasing doses of propranalol on $\frac{dP}{dt}$ max and SW in 4 reserpinized dogs is shown in Figure 18 and table I . There was no reduction in $\frac{dP}{dt}$ max and SW until a cumulative dose of 1.6 mg/kg had been given. Further doses of propranalol depressed the heart. A cumulative dose of 6.4 mg/kg was usually fatal.

In each of the four reserpinized dogs, propranaloldid not increase any of the duration parameters (table I).The maximum change observed in these measurements was a decrease of 13 ms which was observed in the ECG of one animal.

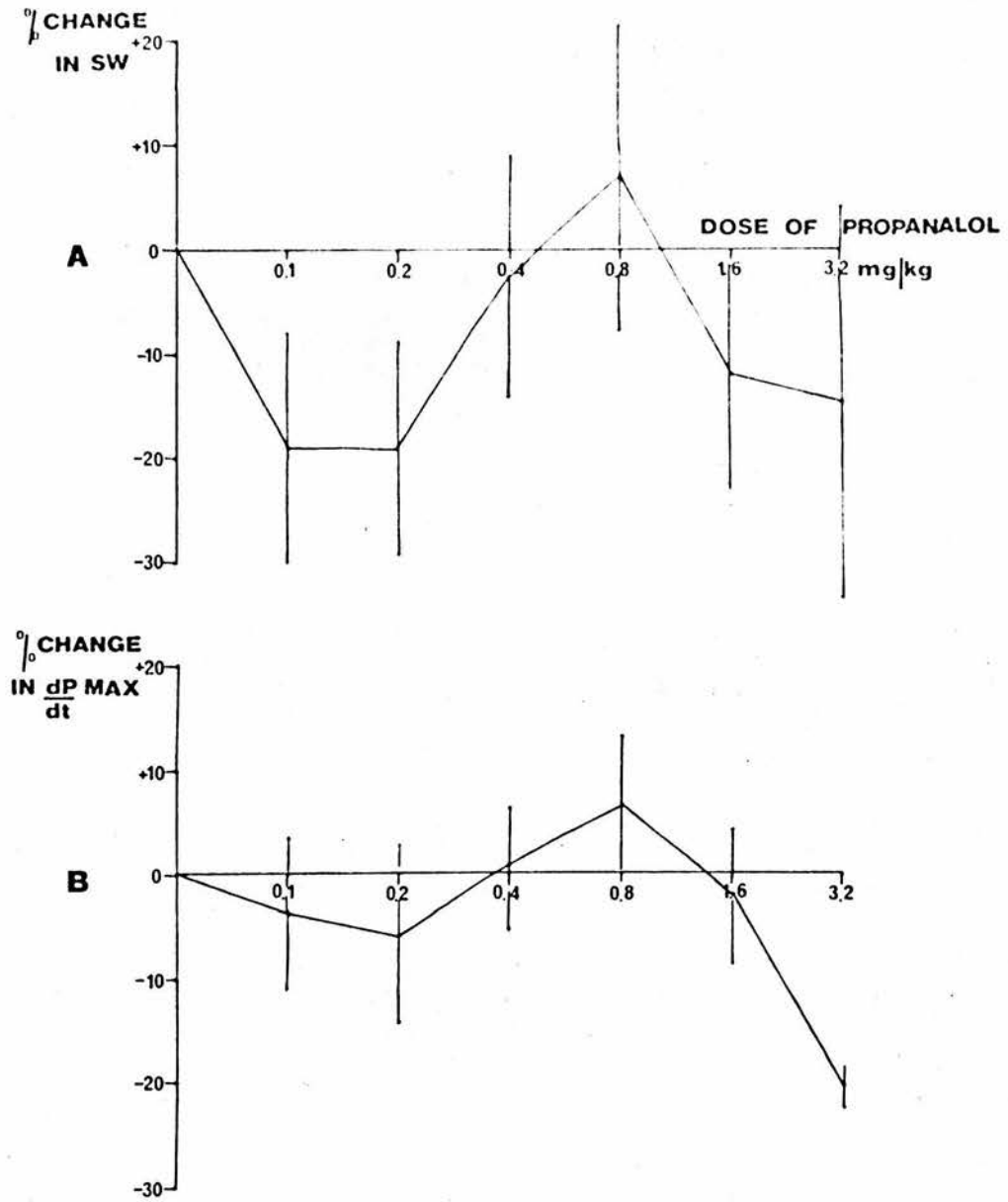


Figure 18. The effect of propranalol on 4 reserpinized dogs, in which HR, MAP and LVEDP were held constant.

A. The relationship between the total does of propranalol and the % change from control in SW $\pm$ 1SEM.

B. The relationship between the total dose of propranalol and the % change from control in $\frac{dP}{dt}$ max $\pm$ 1SEM.

The effect of sotalol after practalol

We decided to repeat the experiments using sotalol, after B adrenergic blockade of the heart had been achieved with practalol, 10 mg/kg, because the reserpinized experiments had been difficult to control satisfactorily and also because we wanted to exclude depression to B blockade of circulating catecholamines observed in both normal and reserpinized dogs. We found this preparation to be very stable and MAP could easily be controlled.

Doses of sotalol below 4 mg/kg had no effect on $\frac{dP}{dt}$ max and SW recorded after the administration of practalol. Further doses produced a small negative inotropic effect, the maximum decrease being 10% at a dose level of 16 mg/kg (Figure 19 table J).

Figure 20 and table J shows the effect of sotalol on the duration parameters in these three animals. The increases in all three parameters were dose-dependent. The most noticeable difference between these results and those obtained in normal dogs is that the increases in all three parameters, at each dose level, are reduced. This effect was most prominent in the measurements of the ECG.

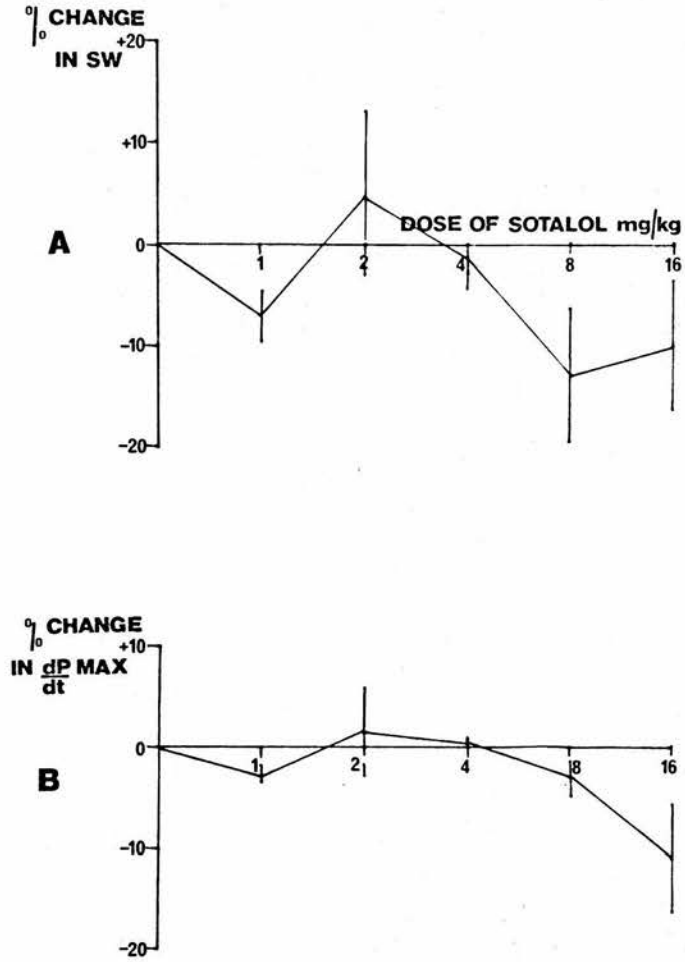


Figure 19. The effect of cumulative doses of sotalol in 3 dogs, which had received practalol. HR, MAP and LVEDP were held constant.

A. The relationship between the total dose of sotalol and the % change from control in SW $\pm$ 1SEM.

B. The relationship between the total dose of sotalol and the % change from control in $\frac{dP}{dt}$ max $\pm$ 1SEM

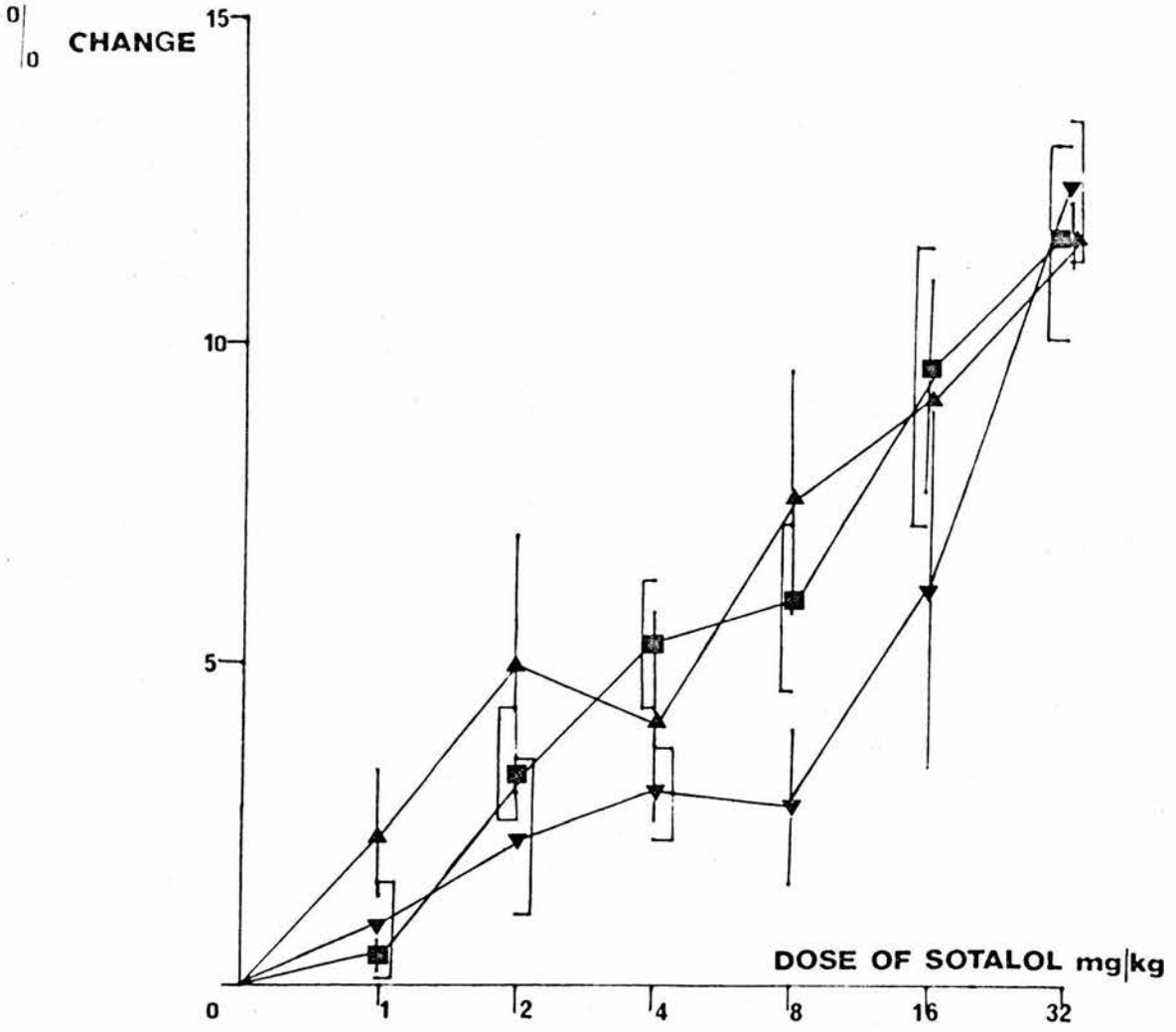


Figure 20. The effect of cumulative doses of sotalol, in dogs which had received practalol, onthe % change in the duration parameters. The mean control duration of the ECG, ▲, was 231 ms(SD± 16), of the ventricular pressure pulse, ■, was 272 ms (SD± 5), and of systole, ▼, was 213 ms (SD± 7).

DISCUSSION:

Sotalol and propranalol are members of a series of drugs which are specific competitive antagonists against the action of sympathomimetic amines on the B receptors in the heart. Evidence from experiments on the myocardial depression exhibited by these drugs has led to conflicting conclusions about the relative effects which result from B blockade and also a non-specific direct depressant action not related to the adrenergic blocking properties of these compounds. Nayler (1970) states that the negative inotropic effect of propranalol at a B blocking concentration of 0.2 mg/kg, cannot be explained by B blockade alone. Harry et al (1973), however, state that in doses below 1 mg/kg propranalol is devoid of a directly depressant action on the heart. Sotalol on the other hand, has been reported to have no direct negative inotropic effect, at B blocking doses (Lish et al 1965). Furthermore, Kaumann and Blinks (1967) and Parmley et al (1972) reported that sotalol had a positive inotropic action on isolated muscle preparations, which was not due to B adrenoreceptor stimulation.

Many studies of the comparative inotropic actions of B blocking agents on intact hearts are difficult to interpret because effects on the heart due to drug induced changes in HR, MAP and LVEDP have not been eliminated. This preparation enabled us to investigate the direct actions of sotalol and propranalol on $\frac{dP}{dt}$ max and SW without secondary effects due to changes in heart rate, MAP and LVEDP.

Sotalol, 1 - 16 mg/kg, decreased both $\frac{dP}{dt}$ max and SW by a mean of 9% in normal dogs. This depression occured after the initial dose; subsequent doses within this range, did not further depress the myocardial force. This suggests that the initial fall in $\frac{dP}{dt}$ max and in SW was due to blockade of the positive inotropic action of circulating catecholamines. This suggestion is supported by the absence of this initial fall in $\frac{dP}{dt}$ max and in SW in dogs in which the action of circulating catecholamines on the heart had previously been blocked by practalol. In two out of three catecholamine depleted dogs, sotalol did not produce a direct depression of the myocardium. The result from the third reserpinized animal is inconsistant with the findings of the other experiments, as the initial dose of sotalol caused a large fall in $\frac{dP}{dt}$ max and in SW. This effect was probably related to the general instability of the reserpinized preparation rather than a negative inotropic action of sotalol. We conclude, therefore, that in doses below 16 mg/kg, sotalol does not have a direct negative inotropic action on the myocardium. Doses of sotalol above 16 mg/kg did depress the heart in all the animals studied, by a direct negative inotropic action.

Propranalol, 0.1 - 1.6 mg/kg, depressed $\frac{dP}{dt}$ max and SW by 20 - 25% in normal dogs. Since these reductions were not observed in catecholamine depleted animals, these changes must have been due to antagonism of the effects of catecholamines at the cardiac B receptor sites. Propranalol, in doses below 1.6 mg/kg, does not have a direct negative inotropic effect, although at higher

dose levels, it has a marked depressant action.

Previous animal experiments have shown that the ratio of the activities of sotalol and propranalol as B adrenergic blockers is within the range of ½ (Farmer and Levy 1968) to 1/17 (Raper and Wale 1968). Using a dose ratio of 16:1 (sotalol: Propranalol), the responses in normal dogs of equiactive B blocking doses of propranalol(0.5 mg/kg) and sotalol (8 mg/kg) can be compared from Figurel4 and 16 . It can be seen that at equiactive blocking doses,propranalol caused a greater decrease in both $\frac{dP}{dt}$ max and in SW than sotalol.

The other characteristic difference between the actions of sotalol and propranalolwhich we observed was that sotalol caused a greater depression of free heart rate than propranalol. We have no quantitative estimate of this effect as we were primarily interested in observing the actions of sotalol at constant heart rate. In five preliminary experiments, however, we observed that a dose of 2 mg/kg of sotalol reduced the heart rate from a mean of 164 to 134 beats/min, whereas propranalol caused no significant change in heart rate.

A general lack of parallel changes in inotropic and chronotropic properties following sotalol and propranalol has previously been suggested (Puri. and Bing 1969, Gomoll and Braunwald 1973). Gomoll and Braunwald (1973) reported findings similar to ours i.e. that at equiactive B blocking doses, propranalol promoted a greater decrease in both contractile force and in $\frac{dP}{dt}$, while

sotalol produced the more profound slowing of heart rate. Their experiments were undertaken in a dog aortic bypass preparation which was denervated and in which aortic flow and pressure could be controlled. They found that in the non-reserpinized bypass preparation, a 22% (46 beats/min) slowing of HR following propranalol (1 mg/kg) was accompanied by a 26% (490 mm Hg s^{-1}) decrease in $\frac{dP}{dt}$ max. Sotalol (8 mg/kg) however, produced a 44% (94 beats/min) reduction in HR which was associated with an 18% (300 mm Hg s^{-1}) depression in $\frac{dP}{dt}$ max. In the reserpine pretreated dog given propranalol, a 2% (3 beats/min) slowing in HR occured with a 14% (230 mm Hg s^{-1}) decrease in $\frac{dP}{dt}$ max, whereas after sotalol a statistically significant ($P < .01$) 19% (27 beats/min) lowering of HR was accompanied by a 6% (130 mm Hg s^{-1}) reduction in $\frac{dP}{dt}$ max.

Singh and Vaughan Williams (1970) postulated that sotalol's ability to prolong the duration of the action potential and the duration of the contraction might be responsible for its less depressant action on the myocardium.

The results of our experiments support the findings of Strauss et al (1970) and Singh and Vaughan Williams (1970), who showed in dog and cat isolated ventricular fibres, stimulated at a constant cycle length, that sotalol prolonged the duration of the action potential by slowing repolarisation. In our experiments, sotalol increased the duration of the Q-T interval of the ECG in a dose dependant manner in normal dogs (Figure 15). This action of sotalol was also observed in dogs, pretreated with practalol,

although the magnitude of the effect was reduced (Figure 20). Of the two catecholamine depleted dogs, given the complete dose range of sotalol, one showed an increase in the duration of the Q-T interval of the ECG following sotalol. Propranalol, however, did not increase the duration of the Q - T interval of the ECG in any of the experiments, but decreased it in one experiment. The results confirm the findings of Morales-Aguilera and Vaughan Williams (1965) inisolated rabbit atria, which showed that propranalol slightly reduced the duration of the action potential.

The action of sotalol on the duration of the action potential may result from a direct effect of the drug on the electrical properties of the myocardium or from its antaganism of the electrical effects of circulating catecholamines. The fact thatpropranalol,in doses which block the inotropic effects of circulating catecholamines, does not prolong the duration of the ECG, is not itself evidence that the action of sotalol must be more than a blocking action of the electrical effects of catecholamines. We cannot conclude from our results if this action of sotalol persists in catecholamine-depleted dogs. We would have liked to continue the experiments in reserpinized preparations but the relatively high failure rate in these animals and the limitation in the number of dogs which were available to us led us to stop these experiments when it became obvious that a large number of dogs would be needed to obtain a conclusive result. The effect of sotalol after B adrenergic

blockade in the heart had been produced by practalol, however, indicates that although a part of the action of sotalol in normal dogs is due to the antagonism of the electrical effects of catecholamines (the change in the ECG induced by sotalol was smaller after practalol) a direct prolongation of the action potential is also likely. This evidence is not conclusive since practalol itself has been shown to have B stimulant actions (Barrett and Carter 1970).

The changes in the duration of the ECG, after sotalol, were associated with increases in the duration of contraction, as indicated by changes in the duration of the ventricular pressure pulse, and also with increases inthe duration of systole. The larger increases in the duration of contraction than in systole, at each dose level, indicates that the relaxation process, after aortic valve closure, was particularly prolonged. If this prolongation of the contraction were contributing to the lesser negative inotropic effect of sotalol than propranalol, one would have expected to observe a differential effect on SW and $\frac{dP}{dt}$ max. From the mechanical properties of muscle, one would expect that an increase in the duration of the contraction would increase the tension or shortening developed and hence the work done in a contraction without necessarily increasing the rate of development of tension. Our results, however, always showed consistant changes in $\frac{dP}{dt}$ max and in SW. We found no evidence to support a positive inotropic action of sotalol, as indicated by an increase in SW, on the intact heart. In dogs, given practalol, sotalol produced no

change in $\frac{dP}{dt}$ max and in SW until doses greater than 16 mg/kg, which had a negative inotropic action. This implies that although the duration of contraction was increased by sotalol, the effect was either not large enough or not early enough to be reflected by a change in SW.

The failure of sotalol to produce an increase in SW or $\frac{dP}{dt}$ max in dogs given practalol, makes it very unlikely that a direct positive inotropic action of sotalol, mediated by an increase in the duration of contraction, is responsible for the less depressant effect of sotalol that propranalol, as suggested by Singh and Vaughan Williams. It has been postulated that the reason for the differences in haemodynamic response between sotalol and propranalol may be the presence of the depressant properties of propranalol. However, our results on reserpinized dogs have shown that in doses below 1.0 mg/kg propranalol does not possess a direct negative inotropic effect. These results have recently been confirmed by Harry et al (1973). They were investigating the B blocking and inotropic effects of propranalol on the intact dog heart, and found that a dose of 0.05 mg/kg of propranalol decreased control heart rate by 1% although the change in control $\frac{dP}{dt}$ max was - 25%. They also showed that propranalol had a differential blocking effect on the two responses, heart rate and contractility, induced by isoprenaline. For a given change in free heart rate, induced by isoprenaline, $\frac{dP}{dt}$ max was less in the presence of propranalol. In order to explain their results, these authors concluded that in the denervated heart, because the uptake

process for sympathomimetic amines in the myocardium is smaller than that which occurs at the sinu-atrial node (Furnival et al 1971), the concentration of circulating catecholamines in their preparation was not high enough to affect heart rate, but was high enough to have an effect on $\frac{dP}{dt}$ max equivalent to about 25% and it is this effect which is blocked by propranalol. Therefore, propranalolis able to block the effect of endogenous circulating catecholamines only in the myocardial muscle and subsequently there is a fall in $\frac{dP}{dt}$ max with no change in heart rate.

Although this reasoning can explain the experimental results observed with propranalol, it fails to explain the comparative effects of sotalol and propranalol. We have shown that at equiactive B-blocking doses, propranalol promoted greater decreases in contractility, while sotalol produced the greater slowing of heart rate. Moreover Brooks et al (1971) have shown in the intact dog heart that sotalol blocked the isoprenaline induced chronotropic response by 42% although the inotropic response was only slightly decreased. This effect is the opposite to that observed with propranalol (Harry et al 1973). These comparative effects of sotalol and propranalolcannot be explained on the basis of differential uptake of sympathomimetic amines into the sinu-atrial node and myocardial muscle.

The marked effect of sotalol on free heart rate and on heart rate changes induced by isoprenaline may be due partly to a direct effect of sotalol on the electrical properties of the myocardium. As stated previously, we have been unable to conclude from

our experiments whether such an effect exists. The difference in the inotropic responses to sotalol and propranalol cannot be explained by a superimposed positive inotropic effect of sotalol nor by an additional negative inotropic effect of propranalol. The differential responsiveness of heart rate and contractility to B adrenergic blockade raises the possibility of qualitative differences in the receptor sites mediating beta adrenergic responses. Sotalol may be less efficient in blocking B receptors in the myocardium than propranalol: whereas propranlol is less efficient in blocking the B receptors in the sinu-atrial node. From these experiments, however, we can only infer that the differential chronotropic and inotropic responses to sotalol and propranalol, indicates different sub groups of cardiac B receptors.

Clinically there may be potentially significant benefits to be gained from the use of a B blocking compound, like sotalol. Its greater chronotropic blocking action may make it particularly useful as an anti-anginal agent, since heart rate is a major determinant of myocardial oxygen consumption. However, sotalol has less of a negative inotropic action than propranalol, and this would seem to be a particularly important property when it is desired to achieve B blockade in patients with impaired myocardial function. Sotalol's greater chronotropic B adrenoreceptor blocking properties may make it a safer antiarrhythmic for patients with myocardial infarction, in whom a negative inotropic effect would be dangerous.

S E C T I O N 3

R E S U L T S

Haemodynamic actions of bretylium tosylate

The effect of bretylium

1. on the normal dog.

When bretylium was administered to a normal dog, a dose of 0.4 mg/kg, produced a 55% increase in $\frac{dP}{dt}$ max and a 77% increase in SW, measured at constant HR, MAP and LVEDP. With doses greater than 1.6 mg/kg, however, it was no longer possible to maintain the HR at its control value. After a single injection of 10 mg/kg we made no attempt to control the pressures and HR. HR rose from 150 to 208 beats/min MAP initially fell. The fall in MAP, which lasted less than 1 minute, was associated with either an increase or no change in cardiac output and was therefore assumed to be due to peripheral dilatation. It was followed by a rise in MAP from the resting level of 85 mmHg to 140 mm Hg. These changes were accompanied by a very large increase in $\frac{dP}{dt}$ max and SW.

2. On catecholamine depleted dogs.

All these animals appeared lethargic and depressed 24 hours after the administration of reserpine. The initial dose of anaesthetic required was usually reduced to a half of that required

for normal animals. The response to tyramine was measured at the beginning of the experiment to test the degree of depletion of catecholamines in the heart, MAP was increased by a mean of 12% (range 2-29%) and HR was increased by a mean of 1% (range 0-3%). These responses demonstrated that substantial depletion of catecholamines had been produced by the reserpine.

Figure 21 , shows the pooled data from 5 reserpinized dogs. The data from which these results were obtained are tabulated in table K .

The mean HR was 136 beats/min ($SD^{\pm}$ 19), the mean MAP was 76 mm Hg ($SD^{\pm}$ 12), and the mean LVEDP was 1.7 mm Hg ($SD^{\pm}$ 0.7). For each test, HR was held constant, MAP was held to within $\pm$ 4mm Hg of the control valve, except in 3 tests, and LVEDP was held to within $\pm$ 0.4 mm Hg of the control value.

Compared with the responses in the unreserpinized animals the inotropic responses are markedly attenuated and increases in the dose of bretylium above the initial dose do not produce further increases in the inotropic responses. The increases in $\frac{dP}{dt}$ max and in SW produced by bretylium in the reserpinized animals were significant.

Although the pressor responses were also markedly attenuated in the reserpinized animals, the initial vasodilatation was comparable in magnitude in the reserpinized and in the normal dog.

The effect of bretylium on the duration of the ventricular

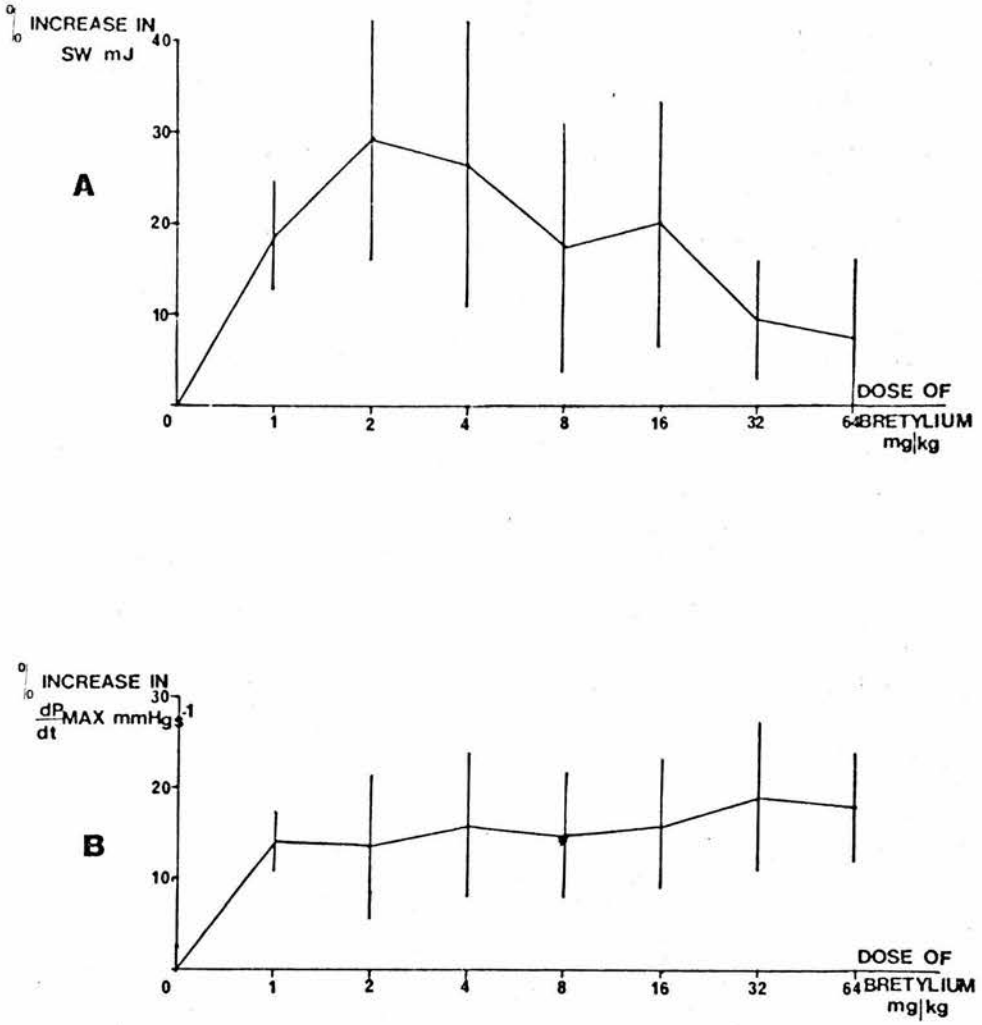


Figure 21. The effect of bretylium on dP max and SW in reserpinized dogs.

A. The relationship between the log total dose of bretylium and the % change in SW from control $\pm$ 1SEM. The mean resting value of SW was 62.4 mJ (SD $\pm$ 8.9).

B. The relationship between the log total dose of bretylium and the % change in $\frac{dP}{dt}$ max from control $\pm$ 1SEM. The mean resting value of $\frac{dP}{dt}$ max was 1340 mm Hgs^{-1} (SD $\pm$ 327).

pressure pulse, the duration of systole and the duration of the ECG are shown in table 2. There was no consistant effect of bretylium on the duration of the ECG, but the positive inotropic effects of bretylium were accompanied in each of the five experiments by a shortening in the duration of systole and of the ventricular pressure pulse. The maximum shortening was of the order of 5 to 11% of the control values.

3. On dogs after blockade of the B_1 receptors in heart.

Bretylium in doses up to 16 mg/kg, when given after practolol, produced no change in $\frac{dP}{dt}$ max and SW measured at constant HR, MAP and LVEDP in 3 dogs. Doses greater than 16 mg/kg reduced $\frac{dP}{dt}$ max and SW (Figure 22). The data from which these results were obtained is tabulated in table L. The mean HR was 161 beats/min (SD$\pm$9), the mean MAP was 85 mm Hg (SD$\pm$2), and the mean LVEDP was 2.6 mm Hg (SD$\pm$ .09). For each test, MAP and LVEDP were held to within $\pm$ 5 mm Hg and $\pm$ 0.5 mm Hg of the control values respectively.

The initial fall in MAP was unchanged from that in the normal animal, but the delayed rise in MAP was smaller than the increase observed before blockade. As the dose of bretylium was increased, the initial fall in MAP became larger and longer in duration. At doses greater than 16 mg/kg, MAP did not fully recover from the initial fall and the delayed increase in MAP did not occur (Figure 23).

The effects of bretylium, after practolol, on the duration of

TABLE 2.

The effect of bretylium on the duration parameters in reserpinized dogs.

Dose of bretylium mg/kg	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of Q - T interval of ECG ms
CONTROL	258	224	246
1	243	206	246
2	256	221	240
4	255	217	241
8	253	216	244
16	258	223	245
32	246	205	234

Figures quoted are the means of five experiments. Full data are tabulated in table K.

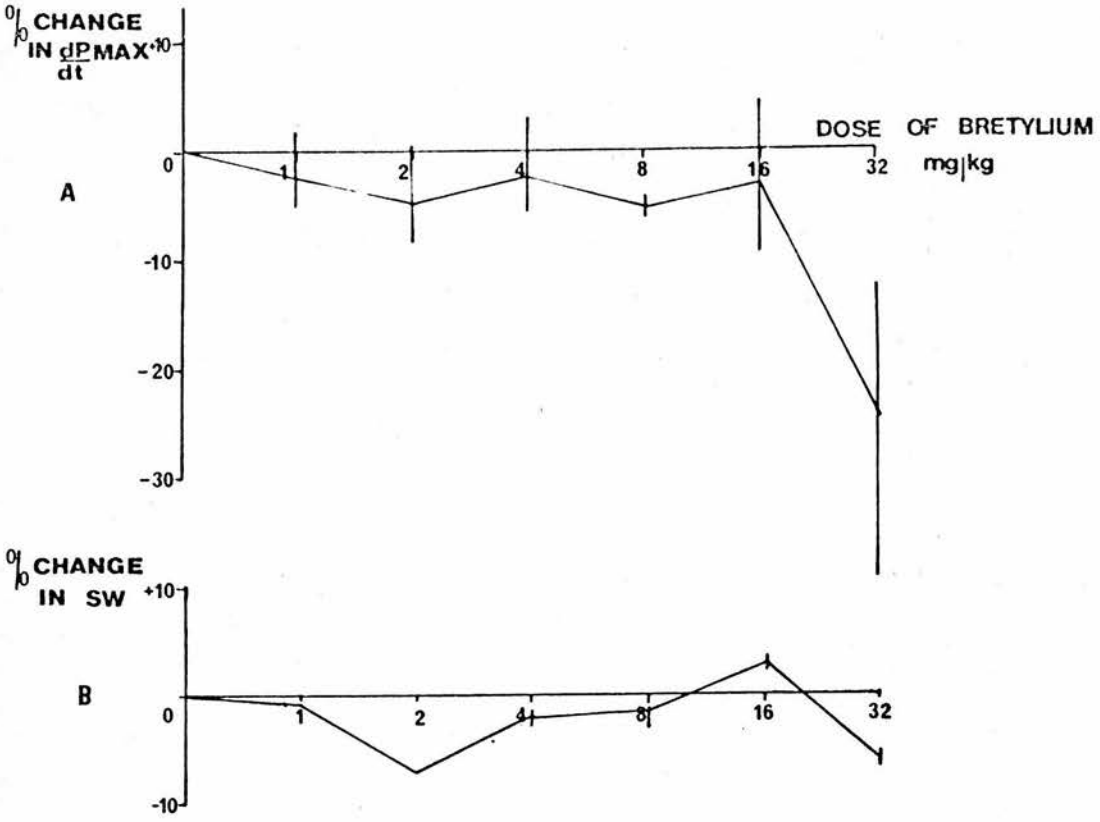


Figure 22. The effect of bretylium on $\frac{dP}{dt}$ max and SW after practolol

A. The relationship between log total dose of bretylium and % change in $\frac{dP}{dt}$ max. The mean control value of $\frac{dP}{dt}$ max was 1644 mm Hgs^{-1} (SD $\pm$ 434).

B. The relationship between log total dose of bretylium and % change in SW. The mean control value of SW was 44.6 mJ (SD $\pm$ 28.9).

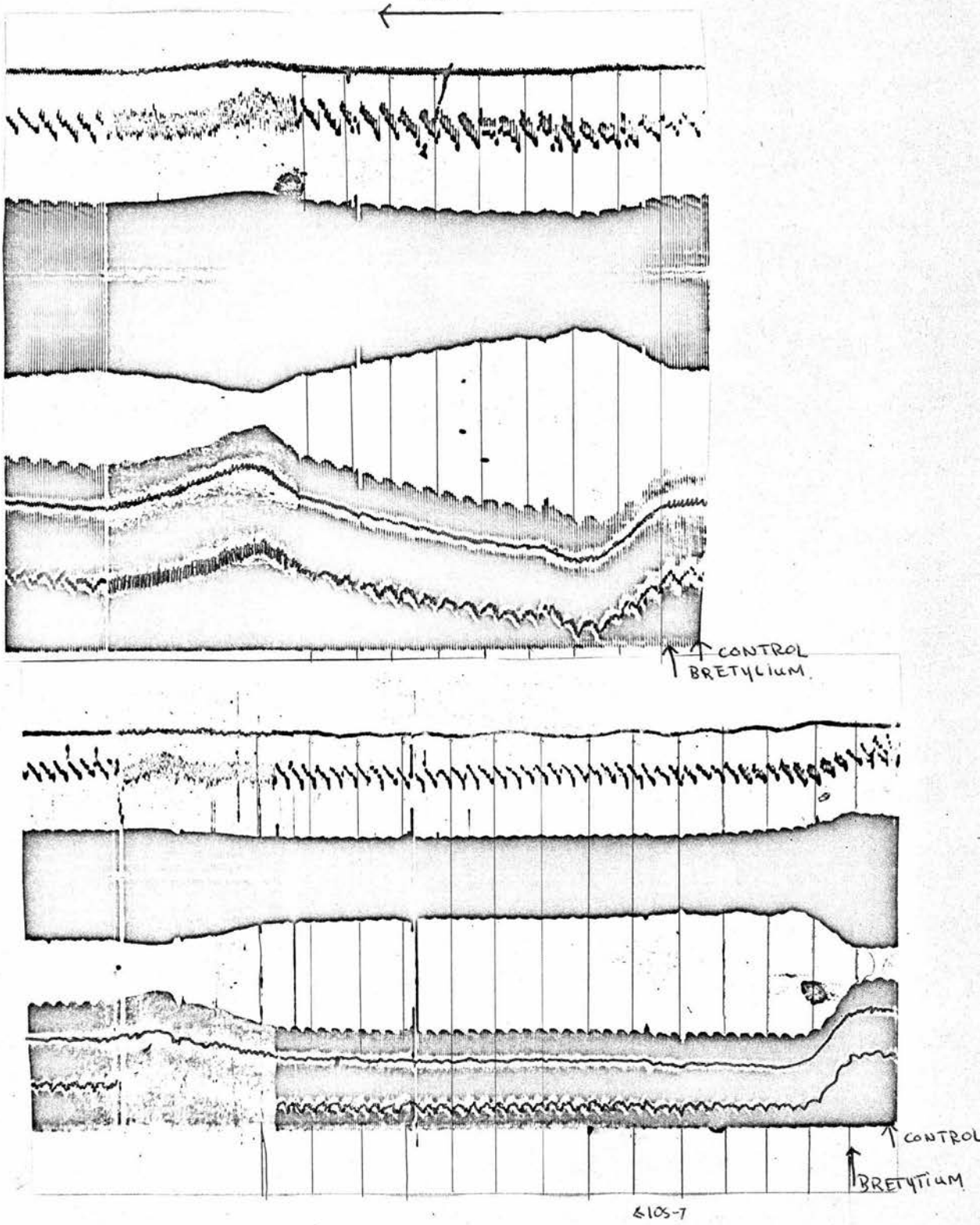


Figure 23. The effect of bretylium on MAP.

A. bretylium, 8 mg/kg

B. bretylium, 16 mg/kg

The records from above downwards are:- C.O., LVEDP, $\frac{dP}{dt}$, LVP, MAP, SW.

the ventricular pressure pulse of systole and of the ECG are shown in table 3. In one experiment out of three, bretylium in doses above 4 mg/kg caused a small increase in the duration of the ECG, and of the ventricular pressure pulse although the duration of systole was unchanged. The maximum increase in duration was of the order of 5% above control values. In the other two experiments there was no change in any of the three duration measurements.

Discussion.

The marked positive inotropic and chronotropic effect of bretylium in the normal dog is associated with its ability to release noradrenaline from adrenergic nerve endings before producing adrenergic blockade (Boura and Green 1959). In order to investigate the direct action of bretylium on the relaxation process in the myocardium, it was therefore necessary to deplete the animals of endogenous catecholamines by prior administration of reserpine. The results of our experiments in reserpinized animals demonstrate that within the range of doses employed, 0.1 to 64 mg/kg, bretylium does not prolong relaxation unless the prolongation is masked by the effect of released catecholamine. Markis and Koch-Weser (1971) were able to show, in reserpinized kitten papillary muscle, a prolongation of relaxation with a concentration of bretylium of $3. 10^{-11}$ M. In normal myocardium they also showed that bretylium did not alter the time required

TABLE 3.

The effect of bretylium on the duration parameters in dogs pretreated with practalol.

Dose of bretylium mg/kg	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of Q - T interval of ECG ms
CONTROL	263	205	181
1	264	204	182
2	264	204	183
4	263	206	184
8	268	210	186
16	275	214	191

Figures quoted are the means of three experiments. Full data are tabulated in table L.

for 90% relaxation. As they concluded that bretylium's positive inotropic action was due to the release of catecholamines, which shorten relaxation time (Koch Weser and Blinks 1963), they also proposed that bretylium had a direct action to slow myocardial relaxation.

This conclusion obtained from isolated muscle studies is not supported by our results. The positive inotropic action of bretylium although much attenuated in reserpinized muscle was not abolished. It was associated with an increase in the rate of development of pressure and a moderate but consistant decrease in the duration of systole. These results suggest that the degree of activation of heart muscle is increased and that the maximum intensity of contraction is achieved sooner in the presence of bretylium. The changes in the time course of contraction seen after administration of bretylium are identical to those produced by noradrenaline. In view of the known catecholamine releasing properties of bretylium, we decided to observe the action of bretylium after the B_1 receptors in the heart had been blocked by practolol. The results of these experiments clearly demonstrate that the positive inotropic effect of bretylium is completely abolished after beta blockade. We conclude, therefore, that the observed positive inotropic effect of bretylium is entirely indirect and is due to the release of catecholamines from their storage sites in the nerve endings in the myocardium. This conclusion supports the earlier findings of Gilmore and Siegel 1962 who demonstrated an increase in coronary

venous catecholamine levels after bretylium administration.

We have found no evidence for a direct positive inotropic action of bretylium. In the one experiment in which we did observe a prolongation of the ventricular pressure pulse, there was no change in the duration of systole. This implies that the observed increase in duration was due to a slowing of the tail of the ventricular pressure curve. Such an effect could not explain the discordant results of Amsterdam et al (1970) and Gaffney and co-workers (1961 and 1962). The results of Amsterdam et al are difficult to explain as the dose of propranalol (10^{-4}M) which they used ought to have been sufficient to produce complete B blockade. The findings of Gaffney and co-workers could be explained if incomplete depletion of myocardial stores of catecholamines by reserpine pretreatment and by cardiac denervation had occurred.

This conclusion implies that the standard regime for reserpinization and the standard test for complete depletion of endogenous catecholamines in the heart are not adequate. We tested the effects of larger doses of tyramine in a dog which had been depleted of catecholamines using the normal dose regime of reserpine. The response to 60 ug/kg of tyramine was measured at the beginning of the experiment as usual. MAP increased by 4% and there was no change in HR. These changes fall within the range of changes observed during the same test in the dogs used to investigate the action of bretylium. Figure 24 shows the effect of higher doses of tyramine in this animal. A dose of

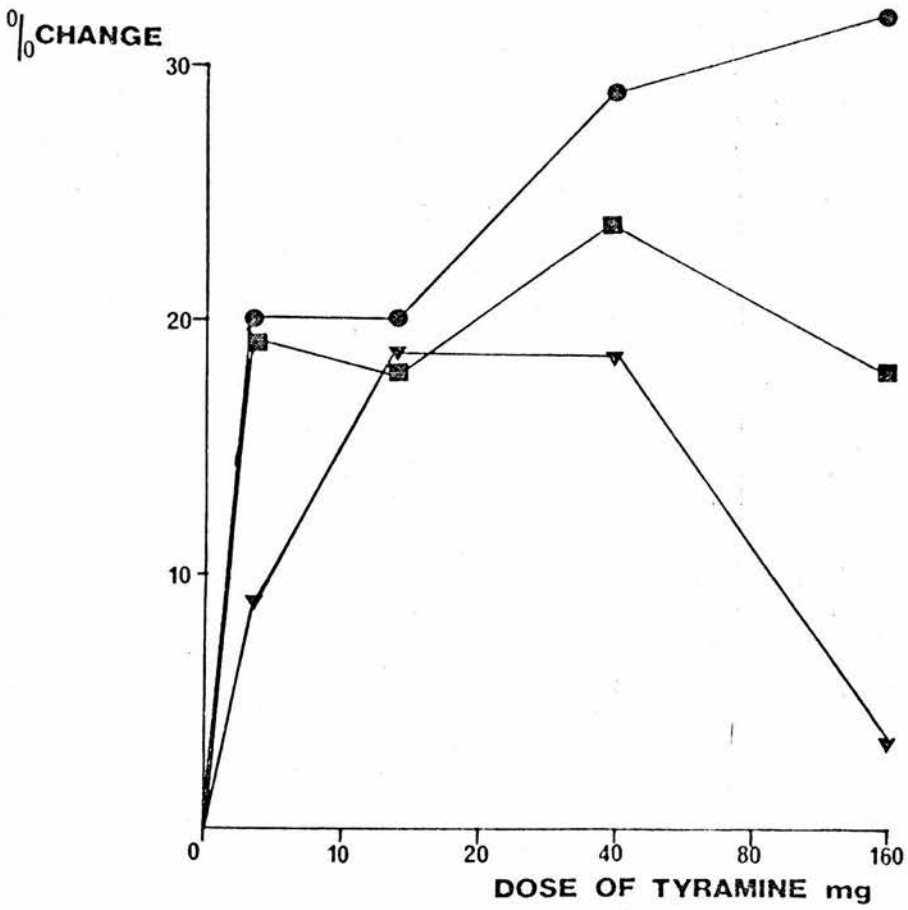


Figure 24. The inotropic and chronotropic effects of high doses of tyramine in one reserpinized dog.

The relationship between the log dose of tyramine and the % change from control in $\frac{dP}{dt}$ max, ■ , SW, ● , and free HR. ▼ . For the measurement of $\frac{dP}{dt}$ max and SW, HR, MAP and LVEDP were held constant, during each test, at the respective values of 121 beats/min 85 mmHg, 2.0 mm Hg.

3.5 mg of tyramine produced a 20% increase in $\frac{dP}{dt}$ max and in SW. Further increases in the dose of tyramine did not produce further increases in the inotropic responses. The positive inotropic effect of tyramine was accompanied in each test by a decrease in the duration of systole and of the ventricular pressure pulse. These doses of tyramine produced a similar response to bretylium in reserpinized hearts. The responses observed were also equal in magnitude. This implies that tyramine and bretylium were acting on the same pool in the nerve endings to release a small fraction of catecholamines which had remained resistant to the depleting action of reserpine. Both intraneuronal (Crout et al 1962) and extra neuronal (Fischer et al 1965) reserpine resistant pools of noradrenaline have been postulated in adrenergic nerve endings, from which tyramine is capable of releasing noradrenaline. Experiments have shown that exposure of reserpine treated tissue to catecholamines can result in the 'refilling' of the pools. Crout et al (1962) for example, found that the response of the reserpinized guinea pig atria to tyramine could be restored to 70% of normal values although the amount of noradrenaline accumulating in the tissue was only 2.2% of the normal level. In reserpinized rat atria, it has also been shown that if doses of bretylium and tyramine, which previously caused no significant inotropic effect, were given after exposure of the atria to noradrenaline, then they both produced a positive inotropic action of the same magnitude which was observed in our experiments (Bhagat and Shideman 1963). The release of catecholamines from the adrenal medulla during induction of anaesthesia or as a result of surgical shock during

our experiments may have resulted in sufficient re-uptake of catecholamines, into the nerve endings, to partially restore the response to tyramine and bretylium. The results of these experiments, therefore, suggest that the lack of an HR change to 60 ug/kg of tyramine is not sufficient to enable one to make the assumption that adequate depletion of catecholamine stores in the myocardium has been achieved. This is especially important when one is investigating the actions of drugs which are powerful releasers of catecholamines.

Bretylium produced an initial peripheral dilatation in all the dogs studied. The magnitude of this depressor response was unaffected by reserpinization. It is probably due to the relatively weaker cholinergic effects of bretylium as previously suggested by Yelnosky and Mortimer (1961). With the higher doses of bretylium the initial depressor response became larger and longer in duration. At doses greater than 8 mg/kg the MAP did not fully recover from this initial depression. This observation may have an important bearing on the clinical use of bretylium, when it is used in these higher doses as an antiarrhythmic agent.

SECTION 4.

The cardiovascular actions of prostaglandin C and E in the cat and dog.

INTRODUCTION:-

Although studies have been made on the actions of the different prostaglandins on the cardiovascular system, these have been mostly limited to investigating their effects on the arteries and veins(Nakano 1973). The inotropic actions of prostaglandins are difficult to evaluate partly because of their potent actions on the peripheral vascular system and also because a considerable species variation in their vascular and cardiac actions limits the extrapolation of experimental data.

Prostaglandin (PG) C has recently been shown to be produced from PGA by an isomerase enzyme present in the plasma of a number of species including the cat and dog (Jones 1972a). The purified prostaglandins C_1 and C_2 were tested for bio-activity in the cat and were found to be several times more potent as a vasodilator than the precursor PGA (Jones 1972b). These experiments also showed that the vasodilator effect of PGC was much more prolonged than that observed with PGE. The prostaglandins C pass through the pulmonary circulation of the cat with negligible loss of vasodepressor activity (Jones and Cammock 1973) unlike the prostaglandins E which are extensively metabolised by the lungs

(Ferreira and Vane 1967). Thus it was thought that this prolonged effect of PGC was simply a reflection of the longer half-life of PGC in the circulation.

Preliminary investigations, however, revealed that following an intravenous infusion of PGC_2 into the cat, a biphasic change in aortic flow occurred, a transient rise followed by a fall, the latter coinciding with the prolonged depression of arterial blood pressure. An example of this effect is shown in Figure 25. We have investigated the secondary fall in aortic flow and its relationship to the prolonged depressor effect to elucidate whether prostaglanins C have actions on the cardiovascular system other than dilatation of the arterioles. We considered three possible actions of PGC.

1. a negative inotropic action on the heart.
2. Contractility depressed by the fall in arterial pressure.
3. A reduction in venous return.

Prostaglandins E have been reported to increase myocardial contractile force in the dog, but not under conditions of constant HR, MAP and LVEDP (Nakano and McCurdy 1967, 1968, Nakano and Cole 1969). We therefore extended our study to the dog and included prostaglandin E for comparison.

The preparation was established as described in the methods section.

RESULTS: The effects of prostaglandins C_1 and C_2, E_1 and E_2 were investigated in the cat.

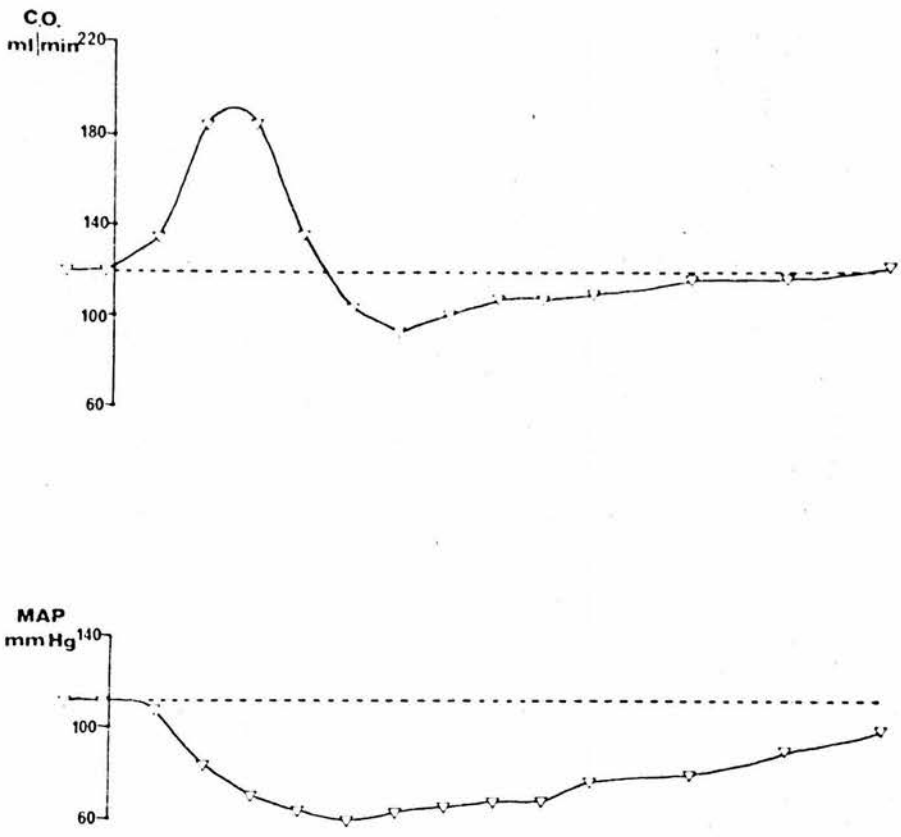


Figure 25. Shows the values of arterial blood pressure and aortic blood flow at 10s intervals of time, after a single intra-aortic injection of 3.5 ug PGC, into a 4.5 kg cat.

Uncontrolled tests in the cat.

Figure26 and tableM shows the results obtained when LVEDP and MAP were allowed to change freely during an infusion of PGC.

In all tests MAP, $\frac{dP}{dt}$ max and SW fell. In 11 out of 13 tests LVEDP also fell. It was not possible to do a parametric analysis of the individual tests because -

a) the resting levels of the parameters varied between experiments.

b) the animals varied in sensitivities to the prostaglandins and

c) in a number of instances long lasting tachyphylaxis to the prostaglandins made it difficult to show dose dependant responses.

However, on a non-parametric basis as judged by the X^2 test the change in LVEDP is significant ($P<0.01$).

Controlled tests in the cat.

The results of the controlled tests for various infusion rates of PGC and PGE are shown in table N , and figure 27 . During each infusion HR was held constant, MAP and LVEDP were held to within $\pm$ 4 mm Hg and $\pm$ 0.3 mm Hg respectively of their control values.

There were no consistant changes in $\frac{dP}{dt}$ max or SW. It can thus be concluded that neither PGC nor PGE has a direct inotropic action on the cat heart.

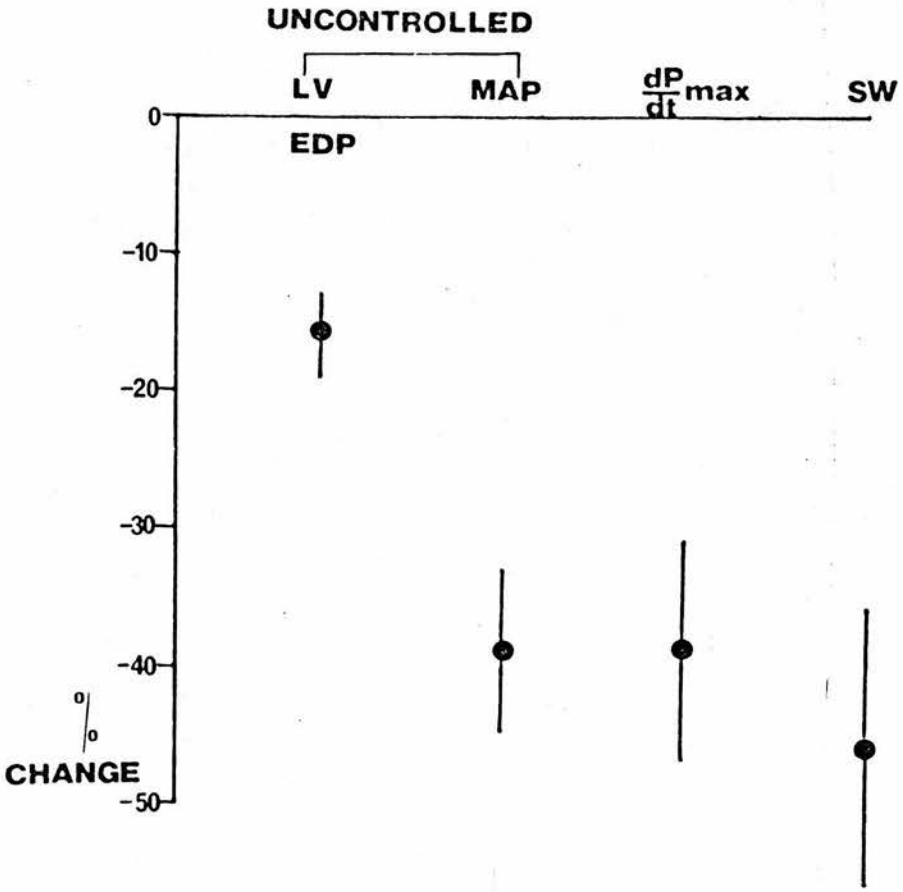


Figure 26. The effect of PGC (0.06 - 5.6 ug/kg/min) on the % change in LVEDP, MAP, $\frac{dP}{dt}$ max and SW, in the cat, when no constraints were applied. The graph shows the means $\pm$ 1SEM for 13 tests in 5 cats. The mean control levels of each parameter are shown in table M.

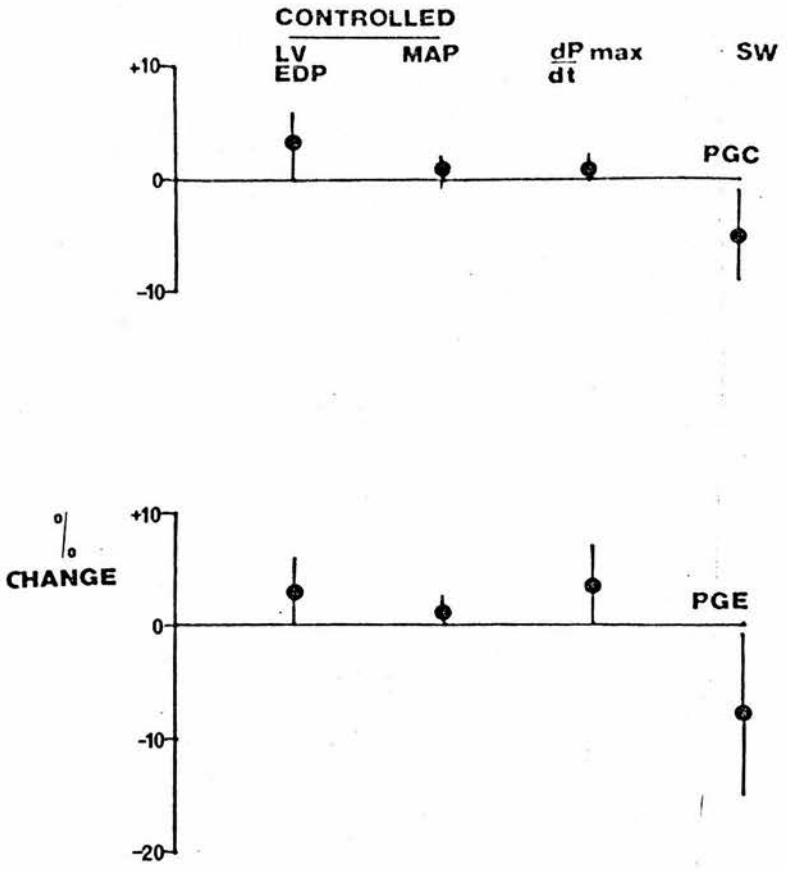


Figure 27. The effects of PGC and PGE on the % change in $\frac{dP}{dt}$ max and SW in the cat, when LVEDP, MAP and HR were maintained at their resting levels. The graphs show the means $\pm$ 1SEM of six tests for each prostaglandin in three cats. The infusion rates varied between 0.06 and 5.6 ug/kg/min for PGC and 0.3 and 2.8 ug/kg/min for PGE. The mean control levels of each parameter are shown in table N.

Controlled tests in the dog.

The effects of PGC_2 and E_2 were investigated in the dog. Heart rate was held constant, MAP and LVEDP were held constant to within $\pm$ 3 mm Hg and $\pm$ 0.3 mm Hg respectively. An increase in $\frac{dP}{dt}$ max and SW occurred in each test, indicating a positive inotropic action on the myocardium. The results are shown in table 0 and figure 28 . The changes in MAP and LVEDP were not significant whereas $\frac{dP}{dt}$ max and SW increased significantly ($P<0.01$) with both prostaglandins C_2 and E_2.

Uncontrolled tests in the dog.

In some tests we observed the action of PGC_2 and E_2 when constraints on MAP and LVEDP were not applied(Figure 29 and table P).With both prostaglandins, MAP fell, SW was unchanged although $\frac{dP}{dt}$ max was significantly increased ($P<0.01$).

DISCUSSION

We have shown that doses of PGC which lower the arterial blood pressure of the cat also lowers its LVEDP, $\frac{dP}{dt}$ max and SW. The effect on $\frac{dP}{dt}$ max and SW is not seen in the paced heart if both LVEDP and MAP are held constant. This is also true with PGE. We conclude, therefore, that these prostaglandins have no direct inotropic action on the cat heart. The explanation

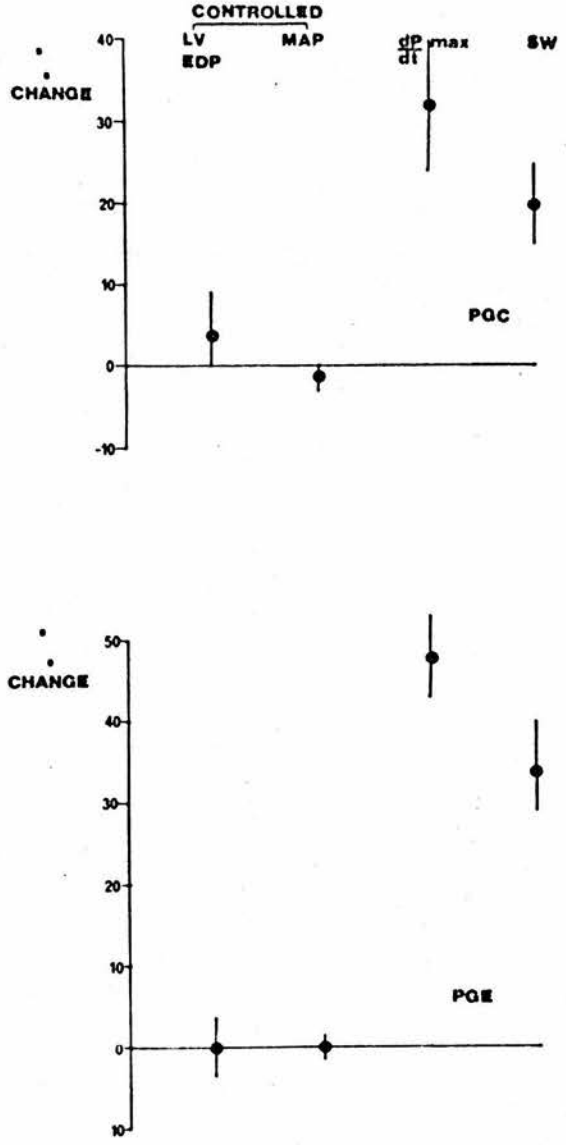


Figure 28. The effects of PGC_2 and PGE_2 (0.07-1 ug/kg/min) on the %changes in $\frac{dP}{dt}$ max and SW of the dog heart, when LVEDP, MAP and HR were maintained at their resting levels. The figure shows the means $\pm$ 1SEM of 4 tests with PGC_2 and 6 tests with PGE_2 in three dogs. The mean control values of each parameter are shown in table 0.

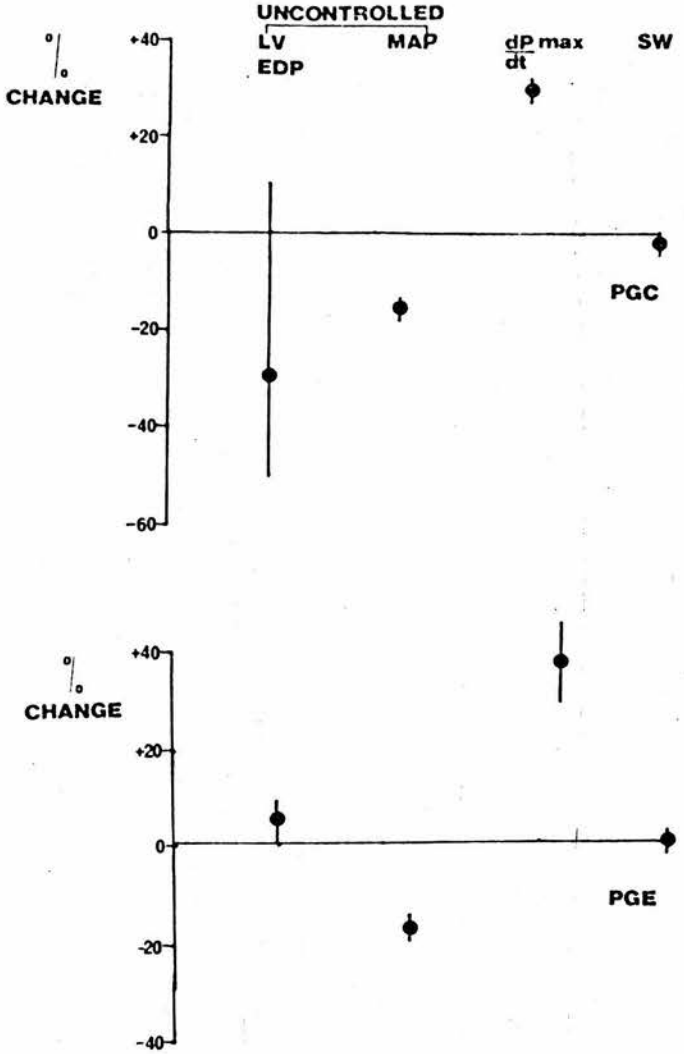


Figure 29, The effects of PGC_2 and PGE_2 (0.07 - 1 ug/kg/min) on the % changes in $\frac{dP}{dt}$ max and SW, in the dog, when no constraints were applied. The figure shows the means $\pm$ 1SEM of 2 tests with PGC_2 and 5 tests with PGE_2 in two dogs. The mean control values of each parameter are shown in table P.

for the fall in aortic flow seen in our preliminary experiments with PGC must, therefore, lie in an action on the peripheral circulation. The fall in $\frac{dP}{dt}$ max and SW, in the uncontrolled state, can be accounted for partly by the fall in arterial pressure and partly by the fall in LVEDP. The fall in arterial pressure is associated with the known potent vasodilator properties of PGC (Jones 1972b). The fall in LVEDP in conjunction with a fall in cardiac output indicates a reduced venous return. The mechanism of this reduction in venous return is not known.

Von Euler (1939) found that a crude PG extract increased portal venous pressure and caused blanching of the liver. Pooling of blood in the portal circulation, due to constriction of vessels in the liver, would reduce venous return. Another possibility is that PGC may dilate capacitance vessels. This could be a direct action on smooth muscle, or an inhibition of adrenergic venomotor tone. Death from circulatory collapse which is seen with large doses of PGC in the cat can be explained by the combination of vasodilatation and reduced venous return.

In contrast with their actions in the cat, PGC_2 and PGE_2 exert positive inotropic actions in the dog. The two compounds are of the same order of potency. These results confirm the findings of Nakano and McCurdy (1967) who showed that PGE_1 increased myocardial contractile force in dogs. Since our measurements were made at constant HR, MAP and LVEDP, the changes in $\frac{dP}{dt}$ max and SW seen in Figure 28 indicate the extent of the direct inotropic action in the absence of effects secondary to peripheral vascular

actions of the prostaglandins.

The mechanism by which PGE and PGC exert their inotropic effect in the dog is not known. The cardiac effects of these prostaglandins cannot be mediated by reflex sympathetic stimulation of the heart as a consequence of the decreased systemic arterial pressure, since in these experiments the heart was denervated. Nakano and McCurdy (1967) also showed that the inotropic effect of PGE was present in dogs pretreated with 1mg/kg of propranalol a dose which completely blocks the cardiac effects of released catecholamines. It has also been suggested that the inotropic actions of protaglandins are secondary to a direct relaxation of coronary vessels. Nutter and Crumly (1972) observed that PGE_1 and PGA_1 dilated coronary vessels after B adrenergic blockade. They also showed, however, that PGA had the largest inotropic action but had less coronary dilator effect than that of PGE_1. Their results therefore, do not support the hypothesis that the cardiac affects of prostaglandins are secondary to coronary dilatation.

The mechanism of the inotropic action of PGE_1 has been studied in the isolated perfused hearts of frog and rat , with similar results in both preparations. The inotropic action is enhanced by a high K^+/Ca^{++} ratio (Vergroessen and de Boer 1968). Hearts arrested by a high potassium concentration can be restored to rhythmic contractility by PGE_1 but only in the presence of an adequate calcium concentration (Piccinini et al 1969). It seems possible, therefore, that the action of PGE_1 on the heart may be

related to an action on calcium influx.

An interesting observation from the uncontrolled tests in the dog should be noted. Figure 29 shows that divergent effects on $\frac{dP}{dt}$ max and on SW have been produced because of the combination of the klinotropic and the vasodilator actions of prostaglandins. Thus although $\frac{dP}{dt}$ max is still increased, in the uncontrolled state, there is no change in SW.

GENERAL DISCUSSION

In recent years much research has been done in an attempt to quantitate the contractile state of the heart and in particular to find a single index that will reflect changes in myocardial contractility. A change in contractility is usually defined as a change in the performance of the heart that arises from changes in the relationship during the 'active state' among force, velocity, fibre length and time after excitation. By definition, therefore, changes in performance arising simply from changes in physical conditions outside the contractile system would not constitute a change in contractility. For this reason, the search for an index of contractility has been centred around those which may be independent of preload and afterload. If as defined it is assumed that changing the afterload does not alter contractility, events in the isovolumic phase of contraction ought to be unaffected by changes in afterload. Attention has therefore been focused on indices derived from this phase of ventricular contraction, particularly those which reflect the velocity of shortening of the contractile element, e.g. $\frac{dP}{dt}$ max. We have shown, however, that $\frac{dP}{dt}$ max is not independent of changes in LVEDP and MAP over the range which we studied. These results confirm previouswork from which it has been concluded that $\frac{dP}{dt}$ max provides a means of detecting alterations in the contractile properties of the heart, only if haemodynamic variables, such as HR, MAP and LVEDP do not change.

As a result of these findings, most of the recent work has been done to try and find indices derived from $\frac{dP}{dt}$ max which correct for changes in preload and afterload (Davidson et al 1974).

We would propose that this line of research has only a limited application because the effects of an intervention on the following three features must be noted before a complete characterisation of an inotropic response can be made.

1. rate of rise of active state
2. peak active state.
3. duration of active state.

Obviously any one index will not describe the overall contractility of an isolated segment of muscle or the whole heart but indices such as $\frac{dP}{dt}$ max are used to reflect changes in contractility because the majority of inotropic interventions alter the speed of contraction and the force of contraction in parallel and also because the importance of changes in the duration of contraction are often not realised. We attempted to show that situations do occur in which the duration of contraction can determine the strength of contraction or in which force and velocity of contraction are changed in different directions such that purely a change in $\frac{dP}{dt}$ max would not reflect the complete inotropic effect.

Work on isolated muscle has shown that tension development or muscle shortening may be limited by an abbreviation of the contraction whereas a prolongation of the contraction increases the

work done either by increasing the time available for tension to develop or for shortening to occur. (Blinks and Koch-Weser 1963). The results of isolated muscle studies suggest that drugs such as sotalol and bretylium produce a positive inotropic effect by prolongation of the duration of the contraction. (Parmley et al 1972: Markis and Koch-Weser 1971). We, therefore, investigated the inotropic actions of these drugs to see if the effect occured in the whole heart and if so, whether such an action would be reflected by a change in SW but not in $\frac{dP}{dt}$ max.

Our results showed that only sotalol increased the duration of the ECG and of the contraction. This effect, however, was not large enough to be reflected as an increase in SW. The results obtained in the whole heart are surprising since the effects on duration in isolated muscle were quite marked. The discrepancy between our results and those obtained in isolated muscle preparations could be due to -

1. a difference in the effective concentration of drug used. The effects of sotalol and bretylium on the relaxation process were observed with concentrations of 10^{-4}M. As we investigated the actions of cumulative doses up to 32 mg/kg, which is approximately 10^{-4}M, it is most unlikely that a difference in dose of drug can explain the discrepancy.

2. a difference in the rate at which the isolated and whole heart muscle were paced. Because of the problem of ensuring adequate oxygenation of the tissue, isolated muscle is usually paced at

rates below 30/min, whereas our hearts were paced about 140 beats/min. Blinks et al (1972) have shown that the prolongation of the contraction observed with the methylxanthines is highly dependant on the frequency of contraction. The effect of caffeine on the duration of contraction and the developed tension was decreased as the interval between contractions was reduced from 300 to 3 seconds. Similar studies have not been done with sotalol and bretylium, however, it seems likely that their action on the relaxation process may be reduced in the whole heart as a result of the higher frequency of stimulation.

Although we have been unable to show the importance of drug induced increases in the duration of contraction, there is much evidence in the literature to support our conclusion that changes in the duration of contraction can produce divergent effects on parameters which measure velocity of contraction and the amount of work done in a contraction.

With the induction of acute hypoxia, for example, a decrease in contractility is observed without a change in the speed of contraction. Tyberg et al (1970) determined Vmax and Po from the force-velocity relationship, in isometric contractions of cat papillary muscle subjected to hypoxia and subsequent reoxygenation. Initially Po fell to 69% of control values, although Vmax had decreased only slightly. The fall in Po was accompanied by a decrease in the duration of contraction. After 60 minutes of hypoxia, Vmax had also fallen to 46% of control values. During early reoxygenation, Vmax increased only slightly from its level

following hypoxia: whereas the duration of contraction became very prolonged, resulting in the restoration of force development to 76% of the control value. The authors suggested, therefore, that the changes in the force of contraction involved the interaction of two effects, with the onset of hypoxia, the duration of 'active state' was abbreviated whereas the intensity (i.e. the rate of development) of 'active state' only declined later. With recovery the duration of 'active state' was immediately prolonged whereas the intensity returned to normal slowly. Similar results have been reported in the intact dog ventricle. (Benzing et al 1974). They found that if myocardial ischaemia was produced by interruption of coronary flow then mean left ventricular power was significantly reduced to 50% of the control values whereas Vmax was still 95% of its control value. This study did not include data of the corresponding effects on the duration of contraction and so it is not possible to say whether these changes in the intact heart were accompanied by similar changes in the duration of contraction observed in the isolated muscle studies.

These results from isolated muscle and the intact heart do show, however, that hypoxia effectively reduces the number of tension generating sites, possibly by reducing the time available for activation, although the speed with which each individual unit contracts is unaffected.

The effects of changes in temperature on muscle function provide another example of a situation in which duration effects are of great importance. Kruta (1937, 1938) has made a detailed

study of the inotropic effects of a change in temperature in isolated guinea pig atria. His results show that a decrease in temperature, at a constant frequency, decreases the rate of development of tension in an isometric contraction but increases the strength and duration of contraction. These results have been confirmed in cat papillary muscle. Cooling from 37^{o} to $19.5^{o}C$ increases the duration of contraction five fold: this effect being associated with a prolongation of the action potential (Trautwien and Dudel 1954). The positive inotropic effect of a decrease in temperature has also been confirmed in the heart in situ (Cotten and Brown 1957). It is postulated that the change in contractile force of the cooled myocardium (which is in the opposite direction that one might expect from the known temperature dependance of the high energy phosphate metabolism) is mainly due to the increased ca^{++} influx into the myocardium which occurs as a result of the prolongation of the action potential (Kaufmann and Fleckenstein 1965).

The results of our experiments provide another two examples of situations in which force and velocity may not be changed in parallel. Increments in heart rate increased $\frac{dP}{dt}$ max although peak systolic pressure was unchanged. This reflects that the speed of contraction was altered but the force of contraction was unchanged. A similar effect was observed if noradrenaline was administered when the heart was paced at a very fast rate. In this situation, the heart was probably already working at the upper limit of its contractile strength and the duration of contraction was so much

reduced that noradrenaline was only able to increase $\frac{dP}{dt}$ max and not peak systolic pressure and hence SW.

The examples given above show that any one derivation from the force-velocity-length-time relations will be inadequate to fit all circumstances. In such situations assumptions are not warranted despite the fact that simple contractile indices may be useful in a given limited circumstance. As has been shown it is not always true to say that an increase in $\frac{dP}{dt}$ max represents a positive inotropic effect. The existing terminology used in studies of cardiac function does not provide for such situations, because the various facets of myocardial contractility have been lumped together and designated by this single term.

This way of thinking can lead to confusion and conflicting reports. This is exemplified by the experiments which have been done to assess myocardial and circulatory effects of bacterial endotoxin. Goodyer (1967) who used $\frac{dP}{dt}$ max as an index of contractility concluded that, in the dog, ventricular contractility was maintained after endotoxin. In contrast to these findings, Solis and Downing (1966), in a modified areflexic cat heart-lung preparation, showed that following endotoxin administration there was a progressive myocardial deterioration, as indicated by a decrease in stroke volume at constant LVEDP, HR and MAP. In order to explain the discrepancy in the results, Parratt (1973) repeated these studies in an intact cat preparation, in which simultaneous measurements of $\frac{dP}{dt}$ max and cardiac output were made. He showed that after endotoxin, HR and $\frac{dP}{dt}$ max were increased,

LVEDP was unchanged but MAP and SW were considerably reduced. The author concluded that because there was a rise in $\frac{dP}{dt}$ max, myocardial contractility had been maintained by increased sympathetic discharge. This conclusion was made in spite of the fact that cardiac output and SW were reduced as a consequence of which the animals died in circulatory failure. A similar effect was seen in our experiments involving the action of prostagladins in the dog. Under uncontrolled conditions, prostaglandins increased $\frac{dP}{dt}$ max although SW was unchanged. In these cases it is not possible to equate changes in $\frac{dP}{dt}$ max with changes in the performance of the heart as a pump and, therefore, it is misleading to state that contractility is maintained.

The first step in an effort to classify the cellular mode of action of inotropic interventions must be to distinguish between the various facets of contractility. To do this, one must think of changes in contractility in terms of the fundamental properties of muscle rather than in terms of some index which is only appropriate to the conditions of a particular set of experiments. Furnival et al (1970) used a decrease in the duration of systole as an index of a positive inotropic effect induced by isoprenaline. This is an example of a situation in which an index has been chosen purely to suit the conditions of that particular experiment, because in terms of the mechanical properties of muscle it is wrong to say that a decrease in the duration of contraction represents a positive inotropic effect.

We support Reiter's suggestion (1972) that the term klinotropic be revived to describe changes in the rate of development of force and that inotropic interventions should be classified according to their klinotropic effect and also their effect on the duration of contraction, because an understanding of inotropic effects in terms of these properties seems essential for further progress. It follows directly from this opinion that any single index of contractility will not be universally applicable and therefore we would question whether the goal of developing such an index is an attainable one.

TABLE A.

The effect of changes in HR on $\frac{dP}{dt}$ max, SW and cardiac power.

Heart Rate beats/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	cardiac power watts ÷60
113	1595	80.3	70	2.3	9.07 ÷60
121	1658	77.3	67	2.5	9.35 "
129	1656	64.3	68	2.3	8.29
139	1687	56.7	71	2.4	7.88
150	1722	45.8	69	2.2	6.87
170	1220	39.5	80	5.3	6.71
188	1390	48.9	78	5.0	9.19
208	1366	36.5	79	4.9	6.86
234	1500	38.2	78	5.2	8.94
150	1310	45.1	81	3.6	6.77
163	1405	36.6	80	3.5	5.97
179	1488	36.5	78	3.7	6.53
197	1506	27.4	79	3.5	5.4
208	1937	12.2	80	3.8	2.54
156	1519	78.6	85	1.7	12.26
170	1621	78.3	85	1.4	13.31
188	1637	58.2	83	1.5	10.94
208	1850	58.0	86	1.4	12.06
163	1377	59.4	81	1.6	9.68
179	1719	82.6	82	1.9	14.8
197	1781	62.8	83	1.9	12.4
221	1930	58.3	84	1.8	12.9

TABLE B .

The effect of MAP on $\frac{dP}{dt}$ max and SW.

MAP mm Hg	$\frac{dP}{dt}$ max mm Hg s^{-1}	SW mJ	LVEDP mm Hg	HR beats/min
58	1217	43.5	4.5	150
81	1429	53.3	4.5	150
111	1562	77.7	4.9	150
115	1630	70.1	4.9	150
56	1683	55.1	2.8	139
70	1767	60.7	2.4	139
87	1909	90.9	2.4	139
96	1972	97.2	3.0	139
90	1632	77.7	4.1	156
97	1644	88.7	4.2	156
100	1668	81.1	4.0	156
105	1725	90.7	4.1	156
115	1845	110.1	4.1	156
66	1760	66.7	3.0	163
74	1939	80.2	2.7	163
89	2122	90.8	2.8	163
106	2507	139.8	3.0	163
85	1563	82.4	2.0	156
91	1596	80.7	1.8	156
100	1718	93.4	1.8	156
113	1749	98.3	1.7	156
125	1943	110.7	1.9	156

TABLE C .

The effect of LVEDP on $\frac{dP}{dt}$ max and SW.

LVEDP mm Hg	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	HR beats/min
0.0	941	14.8	82	150
0.8	1145	23.8	81	150
1.5	1301	30.5	83	150
2.6	1364	33.5	89	150
0.9	1422	39.2	75	163
1.6	1645	60.5	78	163
1.9	1702	68.3	80	163
3.6	1786	72.4	79	163
4.9	1793	90.2	81	163
2.5	1130	8.3	61	150
3.3	1232	22.9	63	150
4.8	1335	32.8	64	150
5.8	1381	39.8	65	150
7.2	1428	49.3	66	150
0.6	1455	28.8	77	150
1.6	1558	35.6	75	150
2.5	1787	55.7	78	150
4.2	1698	72.8	76	150
4.8	1858	85.2	79	150
1.0	1341	23.3	83	150
2.6	1368	38.0	80	150
3.5	1511	47.7	80	150
4.2	1468	50.5	80	150
5.1	1561	68.8	84	150
6.0	1586	75.5	83	150
1.6	858	26.1	76	156
3.1	1004	32.3	76	156
3.9	1036	44.4	74	156
4.7	1149	48.0	75	156

TABLE D .

The effect of noradrenaline on dP max, SW and PSP measured at constant MAP, HR and LVEDP.

Rate of infusion of noradrenaline ug/kg/min	$\frac{dP}{dt}$ max $mm\ Hgs^{-1}$	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Conventional SW mJ	Peak systolic pressure mm Hg.
0	1901	26.0	78	2.2	170	25.4	98
0.21	4007	48.2	81	2.0	170	41.4	117
0	1998	27.5	75	2.1	170	27.3	100
0.41	4785	49.1	77	1.9	170	41.5	124
0	1187	51.5	77	3.1	150	50.4	94
0.11	2133	66.7	83	2.9	150	62.1	102
0	1535	52.9	79	4.4	170	55.6	100
0.05	2251	54.8	79	3.9	170	58.4	104
0	1724	35.1	77	1.8	170	26.6	97
0.12	1966	40.5	77	2.1	170	30.7	100
0	1726	31.2	84	1.3	170	25.7	96
0.25	2799	38.2	85	1.5	170	30.5	104
0	2983	36.2	90	2.0	221	32.3	109
0.142	4245	51.9	89	1.8	221	43.8	115
0	2262	31.8	91	0.9	188	20.5	121
0.1	2445	35.2	92	1.3	188	23.2	121
0	1725	48.8	88	2.0	188	35.1	128
0.14	2376	71.8	92	2.0	188	47.7	150
0	1404	36.9	93	2.5	179	32.4	117
0.15	2710	61.8	95	2.0	179	46. 7	133
0	1552	73.5	91	1.8	179	46.1	120
0.05	1759	74.1	87	1.4	179	45.1	122
0	1771	69.8	98	1.1	179	46.9	124
0.1	2689	102.9	98	1.2	179	57.3	143
0	1917	75.9	88	1.0	179	45.6	126
0.2	2714	91.4	89	0.9	179	49.7	143
0	2073	80	86	2.2	208	49.2	109
0.05	2883	125	86	2.5	208	67.5	139
0	2004	75.9	81	1.7	208	45.2	122
0.1	2597	94.2	81	1.6	208	49.6	130
0	1599	63.3	70	2.2	208	41.9	109
0.2	2598	101.7	73	1.9	208	50.5	130
0	1905	92.9	73	2.8	208	62.1	117
0.4	2826	128.6	76	2.4	208	72.8	139
0	2413	58.3	97	1.7	208	38.7	144
0.16	2732	55.5	98	1.9	208	37.8	144
0	2791	76	95	2.5	208	46.7	152
0.32	3747	77	94	2.6	208	45.0	154
0	2601	78.3	96	2.4	208	51.1	148
0.08	2915	86.2	98	2.7	208	52.7	152

Table D . Continued:-

Rate of infusion of noradrenaline ug/kg/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Conventional SW mJ	Peak systolic pressure mm Hg.
0	1762	47.2	81	2.6	179	28.0	124
0.16	2937	75.5	82	2.5	179	39.3	148
0	1674	39.5	79	2.7	179	25.2	128
0.32	3544	84.9	84	2.5	179	41.3	168
0	2075	52.6	85	3.1	179	33.3	116
0.08	2448	64.4	87	3.1	179	37.6	128
0	1631	54.7	68	3.2	139	35.3	112
0.19	2617	85.6	64	3.7	139	45.9	136
0	1926	68.2	81	3.3	179	43.1	132
0.15	2286	66.4	85	3.3	179	42.9	136
0	1896	58.9	82	3.1	179	39.2	128
0.3	2187	52.9	79	3.1	179	35.7	128
0	1929	77.6	118	4.5	150	51.8	141
0.08	2927	90.0	119	4.4	150	58.6	148
0	2188	89	92	2.7	156	37.5	107
0.133	2547	92	92	2.3	156	37.5	107
0	2044	42.2	84	5.6	221	31.3	97
0.133	2800	69.8	88	5.0	221	49.2	107
0	1508	75.8	92	4.1	144	75.9	109
0.107	1901	89.1	92	4.0	144	85.6	110

TABLE E .

The effect of increasing Heart Rate on Peak systolic Pressure and the duration of contraction and systole.

Heart Rate beats/min	duration of contraction ms	duration of systole ms	Peak systolic pressure mm Hg
156	260	217	101
170	253	207	98
188	247	207	104
208	233	193	100
234	210	167	99
113	330	267	117
121	330	273	119
129	323	263	115
139	303	247	112
150	307	248	110
150	285	219	103
163	269	210	100
179	247	193	103
197	230	179	95
208	230	173	91
156	243	200	102
170	230	187	101
188	217	175	97
208	201	167	98
231	205	163	98
163	250	193	106
179	240	187	106
197	230	177	108
221	223	173	106

TABLE F .

Effect of sotalol on normal dogs.

Experiment number	Total dose of sotalol mg/kg	$\frac{dP}{dt}$ max mm Hgs	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats/min	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of ECG ms
K 38	Control	1256	37.8	74	2.6	150	no record on tape		
	1	1180	37.3	75	2.5	150			
	2	1231	42.5	74	2.7	150			
	4	1231	41.9	73	2.7	150			
	8	1396	44.7	75	2.7	150			
	16	1396	49.3	75	2.6	150			
K 46	Control	1982	63.8	81	4.3	179	240	212	197
	1	1584	52.1	80	4.4	179	250	215	210
	2	1549	52.3	81	4.7	179	260	225	208
	4	1473	45.3	79	4.1	179	260	220	213
	8	1548	48.3	79	4.1	179	257	218	223
	16	1528	54.7	78	4.2	179	275	237	220
K 43	Control	1606	34.0	73	2.5	134	295	237	No ECG on record
	1	1526	32.2	71	2.5	134	294	238	
	2	1474	33.2	72	2.9	134	312	239	
	4	1438	27.1	69	2.7	134	312	247	
	8	1544	34.8	75	2.7	134	313	252	
	16	1385	31.3	67	2.4	134	313	249	
K 51	Control	932	56.8	72	5.0	110	310	257	250
	1	909	51.9	70	4.8	110	322	263	283
	2	904	55.9	69	5.0	110	330	258	292
	4	976	59.1	72	5.3	110	327	255	297
	8	918	54.9	72	5.2	110	330	263	294
	16	876	46.9	72	5.3	110	333	270	297

TABLE G .

The effect of propranalol on normal dogs, while HR, MAP & LVEDP are held constant

Experiment number	Dose of Propranalol mg/kg	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Duration of ventricular Pressure ms	Duration of ECG ms
K 41	Control	2591	42.4	83	2.3	188	180	No ECG on record
	0.1	1576	23.1	83	2.6	188	207	
	0.2	2013	36.3	77	2.3	188	209	
	0.4	1851	31.5	73	2.2	188	210	
	0.8	1038	9.9	74	2.0	188	212	
	1.6	1464	16.6	80	1.9	188	237	
	3.2	1475	20.7	77	1.4	188	212	
K 47	Control	1511	63.6	75	2.2	170	228	216
	0.1	1294	44.0	73	2.1	170	232	216
	0.2	1202	29.7	75	2.2	170	224	214
	0.4	1272	30.1	75	2.1	170	232	220
	0.8	1229	27.9	75	2.1	170	230	216
	1.6	1026	23.0	72	2.1	170		
	3.2	929	21.8	72	2.1	170		
K 48	Control	1523	42.7	72	3.2	179	232	208
	0.05	1318	33.4	70	3.2	179	238	208
	0.1	1442	37.6	73	3.1	179	232	208
	0.2	1241	30.5	69	3.2	179	232	208
	0.4	1259	33.1	73	3.3	179	232	209
	0.8	1230	33.5	76	3.3	179	238	200
	1.6	1120	30.4	74	3.2	179	240	200
	3.2	1031	27.8	77	3.5	179	248	208

TABLE H .

The effect of sotalol on reserpinized dogs while HR, MAP & LVEDP are held constant.

Experiment number	Dose of sotalol mg/kg	$\frac{dP}{dt}$ max $mm\ Hgs^{-1}$	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of ECG ms
K 77	Control	1271	56.2	82	2.2	129	267	232	210
	1	1241	55.7	74	2.0	129	267	229	210
	2	1193	52.7	79	2.3	129	273	234	227
	4	1137	45.3	81	2.6	129	273	232	227
	8	1090	40.4	79	2.3	129	280	235	232
	16	1207	52.5	76	2.2	129	284	240	237
	32	1098	42.3	80	2.1	129	282	233	233
K 80	Control	2358	97.6	92	1.8	139	237	212	217
	1	1427	65.6	88	2.1	139	247	217	220
	2	1472	73.0	94	1.8	139	250	213	217
	4	1357	65.7	94	2.0	139	253	215	222
	8	1197	57.8	93	2.0	139	253	218	215
	16	1145	64.2	91	1.9	139	253	217	217
K 86	Control	1290	114.4	65	3.2	121	290	252	285
	0.25	1312	123.6	63	3.0	121	287	252	282
	0.5	1384	128.2	62	2.8	121	287	252	288
	1.0	1366	126.7	61	3.2	121	290	256	287
	2.0	1494	106.6	67	2.6	121	300	252	291

TABLE I .

The effect of propranalol on reserpinized dogs.

Experiment number	Dose of propranalol mg/kg	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of ECG ms
K 81	Control	1409	54.8	78	3.5	179	255	215	220
	0.1	1733	60.2	80	3.7	197	233	202	208
	0.2	1659	55.4	80	3.3	197	238	207	213
	0.4	1518	69.1	78	3.8	179	253	222	225
	0.8	1635	76.9	82	3.2	179	255	223	227
	1.6	1451	55.5	80	3.7	179	255	217	227
K 82	Control	1385	59.2	84	3.3	144	292	227	257
	0.1	1417	55.7	85	3.2	144	287	225	263
	0.2	1388	50.9	84	3.1	144	288	228	260
	0.4	1402	48.7	84	3.5	144	287	220	257
	0.8	1428	46.5	81	3.4	144	287	218	265
	1.6	1306	42.5	81	3.4	144	293	228	262
	3.2	1110	39.1	78	4.3	144	293	232	257
K 83	Control	1454	64.9	84	5.3	170	250	197	192
	0.1	1528	57.0	82	5.4	170	245	193	192
	0.2	1524	61.4	85	6.0	170	250	200	193
	0.4	1593	67.2	79	5.3	170	250	208	193
	0.8	1729	79.6	80	5.2	170	240	200	193
	1.6	1644	70.7	77	5.3	170	247	200	190
	3.2	1709	78.5	86	5.6	170	260	200	233
K 88	Control	1711	43.9	71	2.7	144	258	203	217
	0.1	1393	25.9	73	2.9	144	267	205	218
	0.2	1319	27.1	65	2.8	144	257	202	215
	0.4	1436	33.4	69	2.8	150	258	207	213
	0.8	1529	37.3	74	2.9	150	253	207	203
	1.6	1402	29.9	68	2.8	150	255	205	203
	3.2	1303	29.2	68	2.8	144	263	207	207

TABLE J .

The effect of sotalol on dogs, pretreated with practalol.

Experiment number	Dose of sotalol mg/kg	$\frac{dP}{dt}$ max $mm\ Hgs^{-1}$	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats /min	Duration of ventricular pressure pulse ms	Duration of systole ms	Duration of ECG ms
K 95	Control	2207	68.3	85	1.2	139	274	221	233
	1	2150	64.3	83	1.4	139	277	227	243
	2	2394	80.0	88	1.7	139	289	232	252
	4	2206	65.2	88	1.5	139	293	230	248
	8	2078	59.3	82	1.6	139	298	230	262
	16	1839	50.7	89	1.5	139	313	247	263
K 96	Control	1998	77.8	101	0.5	150	267	203	210
	1	1972	74.9	101	0.6	150	270	203	210
	2	2061	84.3	95	0.7	150	270	207	214
	4	2008	76.7	97	0.6	150	277	207	214
	8	1915	76.4	97	0.6	150	282	212	222
	16	1881	75.0	99	0.6	150	287	221	230
	32	1915	78.1	102	0.6	150	292	225	233
K 98	Control	2453	39.1	94	1.1	150	278	215	249
	1	2336	34.6	93	1.1	150	278	215	256
	3	2289	34.9	93	1.2	150	287	213	266
	8	2462	27.2	92	1.1	150	287	215	263
	16	1970	16.4	81	2.3	150	298	215	263

TABLE K .

The effects of bretylium, measured as constant MAP, LVEDP and HR in reserpinised dogs.

Total dose of bretylium	$\frac{dPmax}{dt}$	SW	MAP	LVEDP	HR	duration of ventricular pressure pulse	duration of systole	duration of ECG
mg/kg	mm Hgs^{-1}	mJ	mm Hg	mm Hg	beats /min.	ms	ms	ms
0	1196	69.8	67	2.7	139	248	no	no ECG
0.1	1256	83.1	67	2.6	139	242	pulsatile	on tape
0.2	1309	92.6	67	3.0	139	235	aortic	
0.4	1469	85.0	69	3.0	139	235	pressure	
0.8	1444	83.3	64	2.9	139	234		
1.6	1471	86.7	68	3.0	139	235		
3.2	1449	84.2	66	3.0	139	235		
6.4	1412	76.1	68	3.0	139	238		
12.8	1404	73.5	64	2.9	139	248		
25.6	1432	82.5	67	2.9	139	245		
0	1600	69.5	80	1.5	150	234	202	no ECG
0.1	1810	73.5	80	1.4	150	236	195	
0.2	1807	72.5	80	1.6	150	233	193	
0.4	1891	72.0	82	1.5	150	230	193	
0.8	2158	104.1	84	1.3	150	220	188	
1.6	2264	117.0	88	1.4	150	218	187	
3.2	2219	103.4	77	1.8	150	223	189	
6.4	2185	90.1	76	1.6	150	219	185	
12.8	2248	103.2	79	1.5	150	216	180	
25.6	2263	105.9	80	1.7	150	220	190	
0	1226	52.5	61	1.3	125	276	228	223
2	1147	51.9	56	1.2	125	271	232	223
4	1365	57.3	58	1.2	125	260	220	221
8	1365	57.9	60	1.2	125	247	217	226
16	1466	60.8	60	1.1	125	270	220	230
32	1417	57.2	64	1.5	125	260	214	222
64	1525	61.4	60	1.1	125	263	217	222
0	1743	52.8	77	1.2	156	271	220	250
1	1642	48.5	74	1.2	156	270	218	250
2	1714	55.0	76	1.5	156	273	218	250
4	1506	39.9	72	0.9	156	273	220	253
8	1629	41.0	76	1.1	156	273	217	253
16	1578	38.4	72	0.9	156	270	215	251
32	1759	48.7	76	1.1	156	260	211	246
64	1945	52.1	76	1.2	156	256	210	243
0	936	67.3	93	2.2	110	293	246	248
1	1074	99.9	88	1.8	110	293	249	243
2	1099	107.8	92	2.1	110	285	246	249
4	1120	116.4	93	2.1	110	283	240	253
8	1083	108.9	91	1.9	110	286	246	253
16	1048	102.8	91	2.0	110	286	240	248

TABLE L .

The effects of bretylium, measured at constant HR, MAP and LVEDP after practolol.

Total dose of Bretylium	$\frac{dP}{dt}$ max	SW	MAP	LVEDP	HR	Ventricular pressure pulse duration	Duration of systole	Duration of ECG.
mg/kg	mm Hgs^{-1}	mJ	mm Hg	mm Hg	beats/min	ms	ms	ms
0	1880	32.2	86	2.5	170	247	203	159
1	1717	-	87	2.7	170	251	205	165
2	1648	-	87	2.3	170	252	205	167
4	1699	31.8	80	2.7	170	247	207	167
8	1767	32.1	83	2.3	170	249	207	167
16	1702	33.3	88	2.7	170	258	213	173
32	1147	30.3	83	2.3	170	-	-	-
0	1143	24.0	81	2.8	170	267	203	185
1	1151	23.3	78	2.5	170	266	198	180
2	1123	20.0	84	2.7	170	266	197	183
4	1155	23.3	79	2.4	170	263	197	184
0	1909	77.6	88	2.6	144	274	209	200
1	1962	78.7	88	2.6	144	276	210	200
2	1947	79.4	93	2.2	144	274	210	200
4	2005	75.6	92	2.1	144	279	213	200
8	1832	75.2	91	2.2	144	287	213	205
16	1997	79.5	88	2.5	144	293	216	210
32	1676	72.6	87	2.5	144	287	208	208

TABLE M .

The effect of PGC in the cat, on $\frac{dP}{dt}$ max and SW when MAP and LVEDP were allowed to change.

Dose of PG ug/kg/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats/min
Control	1859	21.3	91	0.9	208
PGC_2 0.17	1278	13.4	51	0.5	208
Control	1742	15.3	88	2.8	208
PGC_2 3.3	1270	11.1	52	2.0	208
Control	1544	17.3	68	5.4	208
PGC_2 0.06	1091	11.7	43	4.7	208
Control	1379	14.1	73	3.7	208
PGC_2 0.16	1013	8.8	49	3.0	208
Control	2070	19.2	91	3.3	197
PGC_2 0.14	1420	13.0	63	3.1	197
Control	2076	17.8	88	2.4	208
PGC_2 0.28	1564	11.6	68	2.4	208
Control	1525	14.4	67	1.4	188
PGC_2 0.44	1236	9.8	57	1.5	188
Control	3043	48.0	128	2.6	221
PGC_2 0.33	1221	13.8	52	1.9	221
Control	3617	54.9	140	2.1	234
PGC_2 0.06	3319	29.2	127	1.6	234
Control	4577	33.6	135	4.1	163
PGC_1 5.6	2875	20.7	76	3.0	163
Control	3306	28.4	123	4.4	163
PGC_1 5.6	1425	9.4	49	3.5	163
Control	2800	25.2	94	4.6	150
PGC_2 1.7	658	12.2	37	4.3	150
Control	1386	18.5	76	4.2	150
PGC_2 1.7	652	12.7	40	4.1	150
Mean base-line	2379	25.2	97	3.2	193
Mean test	1463	13.6	59	2.7	193
Change +/- SEM	-916 ±184	-11.6 ± 2.5	-38 ± 6	-0.5 ±0.1	- -

TABLE N .

The effect of PGC and PGE on $\frac{dP}{dt}$ max and SW in the cat, while HR, MAP and LVEDP were held constant

Dose of PG ug/kg/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats/min
Control	3043	18.6	121	1.9	234
PGE_2 1.65	3338	19.6	120	2.0	234
Control	2770	28.2	133	1.8	221
PGE_2 0.33	3237	18.0	133	1.7	221
Control	2586	22.7	95	4.2	150
PGE_2 0.83	2654	24.6	95	4.5	150
Control	1644	17.1	82	4.1	150
PGE_2 0.83	1710	18.6	83	4.2	150
Control	3765	33.4	139	4.8	163
PGE_1 2.8	3455	28.4	140	4.7	163
Control	3608	32.5	140	4.9	163
PGE_1 2.8	3601	30.5	141	4.9	163
Mean baseline	2903	25.4	118	3.6	180
Mean test PGE	2999	23.3	119	3.7	180
Change $\pm$ SEM	$\pm$96 $-$99	$-$2.1 $\pm$1.8	$\pm$1 $-$1	$\pm$0.1 $-$0.1	- -
Control	2985	38.7	128	2.1	221
PGC_1 5.6	2985	39.9	132	2.3	221
Control	3300	28.4	123	4.4	163
PGC_1 5.6	3408	23.9	129	4.5	163
Control	2800	25.2	94	4.6	150
PGC_2 1.7	2850	26.5	95	4.7	150
Control	1380	18.5	76	4.2	150
PGC_2 1.7	1374	18.0	76	4.2	150
Control	3043	32.4	128	2.6	221
PGC_2 0.33	3072	26.0	125	2.6	221
Control	3617	36.6	140	2.1	234
PGC_2 0.06	3726	35.6	141	2.2	234
Mean baseline	2854	29.9	115	3.3	190
Mean test PGC	2903	28.3	116	3.4	190
Change $\pm$SEM	+48 $\pm$19	-1.6 $\pm$1.1	+1 $\pm$1	+0.1 $\pm$0.1	- -

TABLE 0 .

The effect of PGC and PGE on $\frac{dP}{dt}$ max and SW, in the dog, while HR, MAP and LVEDP are held constant.

Dose of PG ug/kg/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats/min
Control	1292	38.0	74	3.4	156
PGE_2 1	2117	47.8	71	3.7	156
Control	1080	38.6	68	4.0	156
PGE_2 1	1677	58.3	67	3.7	156
Control	1357	58.1	81	1.6	179
PGE_2 0.28	2133	76.4	79	1.7	179
Control	1628	76.6	85	1.3	179
PGE_2 0.07	2085	82.0	84	1.3	179
Control	1125	31.3	75	2.6	179
PGE_2 0.07	1534	55.7	77	2.6	179
Control	1055	36.2	62	3.3	179
PGE_2 0.28	1636	54.1	65	3.4	179
Mean baseline	1256	46.5	74	2.7	171
Mean test PGE	1864	62.4	74	2.7	171
Change ±SEM	±608 ± 62	±15.9 ± 2.6	- -	- -	- -
Control	1431	52.1	75	3.7	156
PGC_2 1	2148	58.1	73	3.7	156
Control	1708	73.4	83	1.4	179
PGC_2 0.07	2126	81.7	81	1.4	179
Control	977	35.1	61	2.6	179
PGC_2 0.07	1135	41.9	60	2.4	179
Control	1041	32.8	67	1.6	179
PGC_2 0.28	1395	50.4	69	1.9	179
Mean baseline	1289	48.4	72	2.3	173
Mean test PGC	1701	58.0	71	2.4	173
Change ±SEM	±412 -104	±9.6 -2.3	-1 -1	±0.1 -0.1	- -

TABLE P .

The effect of PGC and PGE on $\frac{dP}{dt}$ max and SW, in the dog, when MAP and LVEDP were allowed to change.

Dose of PG ug/kg/min	$\frac{dP}{dt}$ max mm Hgs^{-1}	SW mJ	MAP mm Hg	LVEDP mm Hg	HR beats/min
Control	1638	45.2	73	3.4	156
PGE_2 1	2241	44.5	53	3.7	156
Control	2069	27.7	95	1.6	179
PGE_2 0.28	3276	29.8	80	1.7	179
Control	2414	35.5	100	1.3	179
PGE_2 0.07	3103	33.4	90	1.3	179
Control	1417	64.8	80	1.8	179
PGE_2 0.07	1903	67.4	69	2.3	179
Control	1305	58.3	83	2.5	179
PGE_2 0.28	1602	56.4	65	2.2	179
Mean baseline	1769	46.3	86	2.1	174
Mean test PGE	2425	46.3	71	2.2	174
Change	±656	±0	-15	±0.1	-
±SEM	-136	-0.9	-2.0	-0.1	-
Control	1412	64.1	81	1.3	179
PGC_2 0.07	1712	62.1	70	1.7	179
Control	1262	52.0	81	2.7	179
PGC_2 0.28	1710	52.0	67	1.4	179
Mean baseline	1337	58.0	81	2.0	179
Mean test PGC	1710	57.0	69	1.6	179
Change	±374	-1	-12	-0.4	-
±SEM	-52	-0.7	- 1	-0.6	-

BIBLIOGRAPHY.

Abbot, B.C. and Mommaerts, W.F.H.M., J.Gen. Physiol (1959) 42, 533.

Amsterdam, E.A., Spann, J.F., Mason, D.T. and Zelis, R.F. Am. J. Cardiology, (1970) 25, 81.

Antoni, H., Jacob, R. and Kaufmann, R., Pflugers Arch. (1969) 306, 33

Anrep, G. Von, J. Physiol (1912) 45, 307.

Barrett, A.M., and Carter J., Br. J. Pharmacol (1970). 40, 373.

Benzing, G.,111 Stockert, J., Nave, E., Tsuei, Y.G., and Kaplan,S., Cardiovas . Res. (1974) 8, 313.

Berglund, E., Borst, H.C., Duff, F., and Schreiner, G., Acta Physiol. Scand. (1958) 42, 185.

Bhagat, B., and Shideman, F.E., Brit J. Pharmacol (1963) 20, 56.

Blinks, J.R., Annals N.Y. Acad. Sci (1967) 139, 673

Blinks, J.R., and Koch-Weser, J., J. Pharm. Exp. Therap (1961) 134,373.

Blinks, J.R., and Koch-Weser, J., Pharmacol. Rev. (1963) 15, 531.

Blinks, J.R., Olsen, C.B. Jewell, B.R., and Braveny, P., Circul Res. (1972) 30, 367.

Blix, M., Skand Arch. Physiol (1895) 5, 150

Boura, A.L.A. and Green, A.F., Brit. J. Pharmacol. (1959) 14, 536.

Bowditch, H.P. Arbeits Physiologie (1871) 6, 139.

Brady, A.J., Annals N.Y. Acad. Sci. (1967) 139, 661

Brady, A.J. Phys. Rev. (1968) 48, 570.

Brooks, H. Banas, J.S., Dalen, J.E., and Dexter, L., Am. J. Physiol (1971) 221, 138

Clancy, R.L., Graham, T.P., Ross, J., Sonnenblick, E.H., and Braunwald, E., Am. J. Physiol (1968) 214, 1186.

Cooper, T., Gilbert, J.W. Jr., Bloodwell, R.D., and Crout, J.R., Circul. Res. (1961) 9, 275

Cotten, M. De V. and Brown, T.G. Jr., J. Pharmacol. (1957) 121, 319.

Covell, J.W., Ross, J., Sonnenblick, E.H. and Braunwald, E., Circul. Res. (1966) 19, 364.

Crout, J.R. Muskus, A.J., and Trendelenburg, U., Brit. J. Pharmacol. (1962) 18, 600.

Davidson, D.M., Covell, J.W., Malloch, C.I., and Ross, J., Cardiovas. Res (1974) 8, 299.

De Gubareff, T., and Sleator, W. Jr., J. Pharm. Exp. Therap (1965) 148, 202.

Farmer, J.B., and Levy, G.P., Brit. J. Pharmacol (1968) 34, 116.

Ferreira, S.H., and Vane, J.R., Nat e London (1967) 216, 868.

Fischer, J.E., Kopin, I.J., and Axelrod, J., J Pharm. Exp. Therap. (1965) 147, 181.

Frank, O., Z. Biol. (1895) 32, 370.

Franklin, D.L., Van Citters, R.L., and Rushmer, R.F., Circul. Res. (1962) 11, 702.

Furnival, C.M., Linden, R.J., and Snow, H.M., J. Physiol. (1970) 211, 359.

Furnival, C.M., Linden, R.J., and Snow, H.M., J. Physiol (1971) 214, 15.

Gaffney, T.E., Circul Res. (1961) 9, 83.

Gaffney, T.E., Braunwald, E., and Cooper, T., Circul. Res. (1962) 10, 83.

Gibb, C.L., Circul Res. (1967) Supplement III 21, 223.

Gilmore, J.P., and Siegel, J.H., Circul. Res. (1962) 10, 347.

Gleason, W.L., and Braunwald, E., J. Clin. Invest. (1962) 41, 80

Gomoll, A.W. and Braunwald, E., Arch. Int. Pharmacodyn, (1973) 205, 338.

Goodyer, A.V.N. Am. J. Physiol. (1967) 212, 444.

Hamilton, W.F., Physiol. Rev. (1955) 35, 161.

Hansen, A.T., Acta. Physiol. scand (1950) 19, 17

Hansen, A.T., and Warburg, E., Acta Physiol scand. (1950) 19, 306.

Harry, J.D., Kappagoda, C.T., Linden, R.J., and Snow, H.M., Cardiovas. Res. (1973) 7, 729.

Hefner, L.L., Sheffield, L.T., Cobbs, G.C., and Klip, W., Circul. Res. (1962) 11, 654.

Hill, A.V., Proc. Roy. Soc. London B. (1938) 126, 136.

Hoffman, R.P. and Grupp, G., Dis. chest (1969) 55, 229.

Jones, R.L., J. Lipid Res. (1972a) 13, 511.

Jones, R.L., Brit. J. Pharmacol (1972b) 45, 144P

Jones, R.L., Biochem J. (1974) In the press.

Jones, R.L., and Cammock , S., Advances in Biosciences (1973) 9, 61.

Kaufmann, R. and Fleckenstein, A., Pflugers Archiv. (1965) 285,1

Kaumann, A.J., and Blinks, J.R., Pharmacologist (1967) 9, 248.

Kaumann, A.J. and Olsen, C.B., Science (1968) 161, 293.

Kavaler, F., Am. J. Physiol (1959) 197, 968.

Kavaler, F., and Morad, M., Circul Res. (1966) 18, 492.

Knowlton, F.P., and Starling, E.H., J. Physiol (1912) 44, 206.

Koch-Weser, J. Am. J. Physiol (1963) 204, 451.

Koch-Weser, J., Berlin, C.M., and Blinks, J.R., Proc. IInd Int. Pharmacol. meeting (1963) 5, 63.

Koch-Weser, J. and Blinks, J.R., Pharmacol. Rev. (1963) 15, 601.

Kruta, V., Comptes Rendus Soc. Biolog. (1937) 125, 498.

Kruta, V., Comptes Rendus Soc. Biolog. (1938) 129, 791.

Kruta, V., and Stejskalova, J., Arch. Int. Physiol (1960) 68, 152.

Kruta, V. and Zadina, R., Arch. Pharmacodyn Therap. (1938) 59, 198.

Langer, G.A., Ann. Rev. Physiol (1973) 35, 55.

Lish, P.M., Weikel, J.H. and Duncan, K.W., J. Pharmac. Exp. Therap. (1965) 149, 161.

Markis, J.E., and Koch-Weser, J., J. Pharmac. Exp. Therap. (1971) 178, 94.

Mirsky, I., Ellison, R.C., and Hugenholtz, P.G., Am. J. Cardiol. (1971) 27, 359.

Mitchell, J.H., Wallace, A.G., and Sheldon Skinner, N., Am. J. Physiol (1963) 205, 41.

Morales Aquilera, A., and Vaughan Williams, E.M., Brit. J. Pharmacol. (1965) 24, 332.

Nakano, J., The Prostaglandins Ed. by Cuthbert, M.F., Heinemann, London (1973) Chapt.II, 41.

Nakano, J., and Cole, B., Amer. J. Physiol (1969) 217, 222.

Nakano, J., and McCurdy, J.R., J. Pharmacol, Exp. Therap. (1967) 156, 538

Nakano, J., and McCurdy, J.R., Proc. Roy. Soc. exp. Biol. Med. (1968) 128, 39.

Nayler, W.G., Post grad. Med J. (1970) 46, 90.

Neidergerke, R., J. Physiol (1956) 134, 569.

Noble, M.I.M., Circul. (1972) 45, 252.

Noble, D., and Tsien, R.W., J. Physiol (1969) 200, 205.

Nutter, D.O., and Crumly, H.J., Cardiovas. Res. (1972) 6, 217.

Paasonen, M.K., and Krayer, O., J. Pharm. exp. Therap. (1958) 123, 153.

Parmley, W.W., Rabinowitz, B., Chuck, L., Bonorris, G., and Katz, J.P., J. Clin. Pharm. (1972) 12, 127.

Parratt, J.R., Brit. J. Pharm. (1973) 47, 12.

Piccinini, F., Pomarelli, P., and Chiarra, A., Pharmac. Res. Comm. (1969) 1, 381.

Puri, P.S. and Bing, R.J., Dis. Chest. (1969) 55, 235.

Raper, C., and Wale, J., Eur. J. Pharmac (1968) 4, 1.

Reeves, T.J., Hefner, L.L., Jones, W.B., Coghlan, C., Prieto, G., and Carroll, J., Am. Heart J. (1960) 60, 745.

Reiter, M., Ann. Rev. Pharmac (1972) 12. 111.

Remington, J.W. and Hamilton, W.F., Am. J. Physiol (1947) 150, 292

Reuter, H., J. Physiol (1968) 197, 233.

Rosenbleuth, A., Alanis, J., Lopez, E., and Rubio, R., Arch. Int. Physiol. Biochim (1959b) 67, 358.

Rosenbleuth, A., Alanis, J., Rubio, R., and Lopez, E., Arch Int. Physiol Biochim (1959a) 67, 374.

Rushmer, R.F., Physiol Rev. (1955) 35, 138

Sarnoff, S.J., and Berglund, E., Circulation (1954) 9, 706.

Sarnoff, S.J., Brockman, S.K., Gilmore, J.P. Linden, R.J., and Mitchell, J.H., Circul. Res. (1960a) 8, 1108.

Sarnoff, S.J., Mitchell, J.H., Gilmore, J.P., and Remensnyder, J.P., Circul, Res. (1960b) 8, 1077.

Sarnoff, S.J., Gilmore, J.P. Mitchell, J.H., and Remensnyder, J.P. Am. J. Med. (1963) 34, 440.

Siegel, J.H., and Sonnenblick, E.H., Circul. Res. (1963) 12, 597.

Siegel, J.H., Sonnenblick, E.H., Judge, R.D., and Wilson, W.S. Cardiologia (1964) 45,189.

Siggaard-Anderson, O., Scand J. Clin. Lab. Invest, (1963) 15, 211.

Singh, B.N., and Vaughan Williams, E.M., Brit. J. Pharmac, (1970) 39, 675.

Solis, R.T., and Downing, S.E., Am. J. Physiol (1966) 211, 307.

Sonnenblick, E.H., Am. J. Physiol (1962) 202, 931.

Sonnenblick, E.H., Fed. Proc. (1965) 24, 1396.

Sonnenblick, E.H., and Downing, S.E., Am. J. Physiol (1963) 204, 604.

Starling, E.H., The Linacre Lecture on the Law of the Heart. Cambridge. 1915, London Longmans Green (1918).

Starling, E.H., J. Roy. Army Med. Corps. (1920) 34, 258.

Strauss, H.C., Bigger, J.T., and Hoffman, B.F., Circul, Res. (1970). 26, 661.

Svedmyr, N., Malmberg, R., and Haggendal, E., Pharmacologia Clin. (1970) 2, 82.

Trautwein, W., and Dudel, J. Pflug. Arch. ges. Physiol. (1954) 260, 104.

Tyberg, J.V., Yeatman, L.A., Parmley, W.W., and Sonnenblick, E.H., Am. J. Physiol (1970) 218, 1780.

Vergrossen, A.J., and de Boer, J., Eur. J. P. Pharmac. (1968) 3, 171.

Von Euler, U.S., Scand. Arch. Physiol (1939) 81, 65.

Wallace, A.G., Skinner, N.S., and Mitchell, J.R., Am. J. Physiol (1963) 205, 30.

Weber, A., and Herz, R., J. Gen. Physiol. (1968) 52, 750.

Wiggers, C.J., Am.J. Physiol (1914) 33, 382.

Wiggers, C.J., J. Pharm. exp. Therap (1927) 30, 233.

Wood, E.H., Heddner, R.L., and Weidmann, S., Circul. Res. (1969) 24, 409.

Yelnosky, J. and Mortimer, L.C., Arch. int. Pharmacodyn. Ther. (1961) 130, 200.

ACKNOWLEDGEMENTS

I wish to thank Dr. A. Ungar for supervising this work and also for his kindness and generosity. I greatly appreciate the expert technical assistance of Mr. P. Whelpdale.

I would also like to thank Professor E. W. Horton for allowing me to work in the department and also for his interest in this work.

I am also grateful to the Medical Faculty of Edinburgh University for the scholarship which I was awarded.